

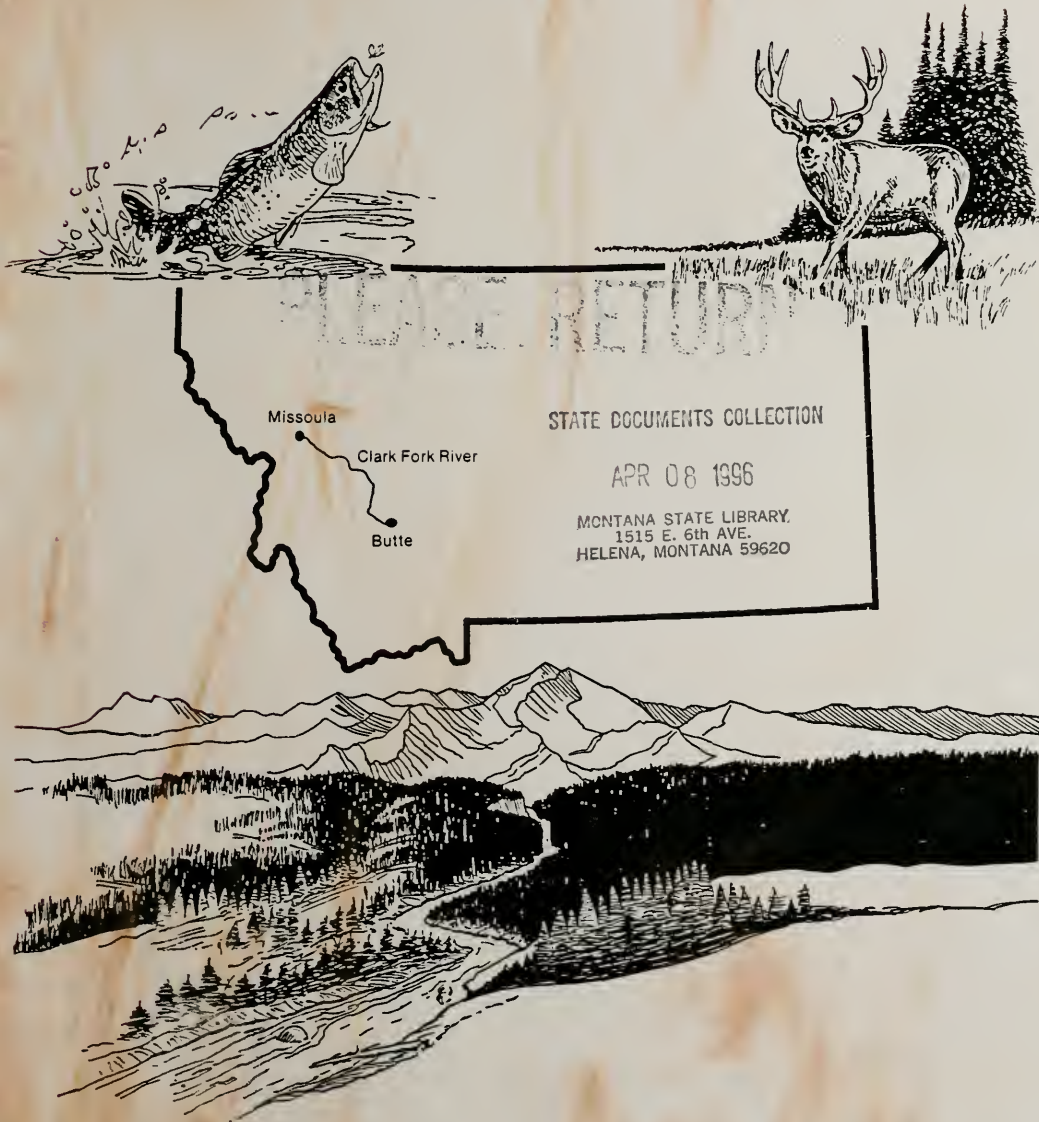
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Expert opinions  
regarding injuries  
to aquatic  
resources, Clark  
Fork River Basin,

STATE OF MONTANA  
NATURAL RESOURCE DAMAGE PROGRAM

EXPERT REBUTTAL OPINIONS REGARDING  
INJURIES TO AQUATIC RESOURCES  
CLARK FORK RIVER BASIN, MT

OCTOBER 1995



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**EXPERT OPINIONS REGARDING  
INJURIES TO AQUATIC RESOURCES,  
CLARK FORK RIVER BASIN, MT**

*Prepared for:*

State of Montana  
Natural Resource Damage Litigation Program

*Prepared by:*

Hagler Bailly Consulting, Inc.  
P.O. Drawer O  
Boulder, CO 80306-1906  
(303) 449-5515

October 18, 1995





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CLARK FORK RIVER BASIN, MT**

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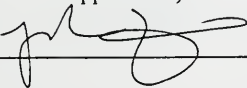
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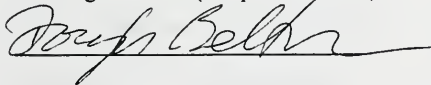
Hagler Bailly Consulting, Inc.  
P.O. Drawer O  
Boulder, CO 80306-1906  
(303) 449-5515

*Testifying Experts:*

Dr. Joshua Lipton (all Chapters, excluding  
appendices)

  
\_\_\_\_\_

Mr. Doug Beltman (Chapters 2 and 3)

  
\_\_\_\_\_

Dr. Harold Bergman, U. Wyoming  
(Chapter 5)

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Harold Bergman

October 18, 1995



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## ACRONYMS

ASTM	American Society for Testing and Materials
BOD	biological oxygen demand
DOC	dissolved organic compound
EDTA	ethylenedinitrilotetraacetic acid
NTA	nitrilotriacetic acid
USGS	United States Geological Survey
WWTP	Wastewater Treatment Plant
WUA	Weighted Usable Area



---

## **CHAPTER 1**

### **INTRODUCTION**

This report, and its related appendices, contains rebuttal opinions regarding injuries to aquatic resources of the Clark Fork River Basin, MT (including surface water and sediments, benthic macroinvertebrates, and fish). These opinions are based on review of reports submitted by the Atlantic Richfield Company (ARCO), review of, and familiarity with issues and literature addressed in ARCO reports, and work performed by experts for the State of Montana. The opinions presented in this report are reliant on opinions of a number of experts for the State of Montana, including:

- ▶ Dr. Harold Bergman, University of Wyoming (Appendices B, C, H, I, Chapter 5)
- ▶ Dr. Richard Brand, University of California/Berkeley (Appendix G)
- ▶ Dr. Don Chapman, Don Chapman Consultants, Inc. (Appendices D and E)
- ▶ Dr. Tracy Hillman, Don Chapman Consultants, Inc. (Appendix E)
- ▶ Mr. Sherman Jensen, White Horse Associates, Inc. (Appendix F)
- ▶ Mr. Mark Kerr, Montana Natural Resource Damages Litigation Program (Appendix A)
- ▶ Dr. Lyman McDonald, WEST, Inc. (Appendix B)
- ▶ Mr. Dan Woodward, National Biological Service (Appendices B, C, I).

This report is organized as follows: Chapter 2.0 contains opinions regarding sediment resources. Chapter 3.0 addresses benthic macroinvertebrates. Chapter 4.0 addresses surface water resources, and relies on the opinions of Mr. Mark Kerr (Appendix A) and Dr. Richard Brand (Appendix G). Chapter 5.0 presents opinions regarding determination of toxicological injuries to fishery resources, and relies on opinions of Dr. Bergman (Appendices B, C, H, I), Mr. Woodward (Appendices B, C, I), Dr. McDonald (Appendix B) as well as Mr. Kerr (Appendix A). Chapter 6.0 presents opinions regarding quantification of injuries to fishery resources, and relies on opinions of Dr. Chapman (Appendices D and E), Dr. Hillman (Appendix E), and Mr. Jensen (Appendix F).



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## CHAPTER 2

### SEDIMENT RESOURCES

#### *2.1 ARCO Contends that Analysis of Fine-Grained Sediments by the State Misrepresents Conditions in the Clark Fork River*

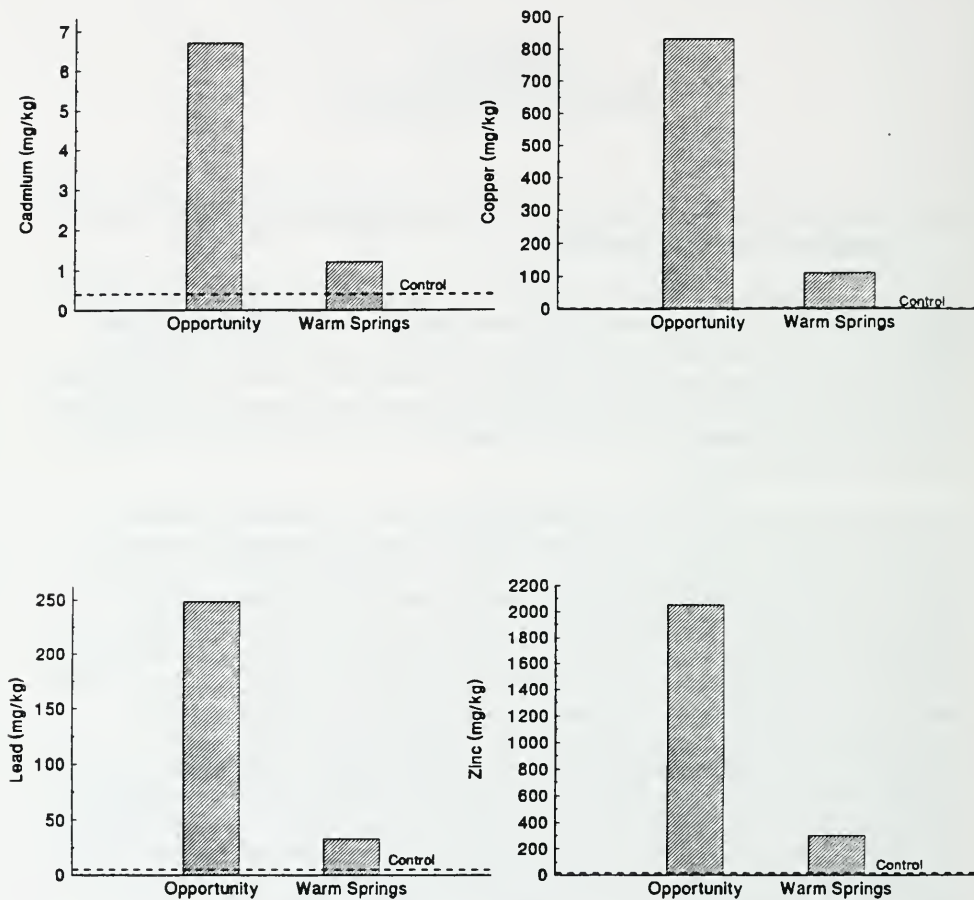
ARCO's reports include a critique of the State's use of fine-grained sediments to characterize hazardous substance concentrations in Silver Bow Creek and the Clark Fork River (Ginn, 1995). ARCO claims that analysis of fine-grained sediments without characterizing bulk sediments "does not provide an accurate portrayal of metals distribution in river sediments or of metals transport in the river" (Ginn, 1995). ARCO states that this approach produces biased results and an "overstatement of the relative contamination of sediments by metals" (Ginn, 1995) in the State's evaluation.

As described in the State's Report of Assessment (Lipton et al., 1995a), fine-grained sediments are a more reliable and less biased method of assessing sediment contamination (Axtmann and Luoma, 1991), and represent an important pathway to biota (e.g., Cain et al., 1992; Luoma, 1992; Gower and Darlington, 1990; Luoma, 1989). Nevertheless, bulk sediments of Silver Bow Creek and the Clark Fork River also are highly contaminated with the hazardous substances arsenic, cadmium, copper, lead, and zinc. For example, Figures 3-7 through 3-11 of the State's Report of Assessment demonstrate that arsenic, cadmium, copper, lead, and zinc concentrations in bulk sediments of Silver Bow Creek are well above concentrations in control stream sediments<sup>1</sup>. More recent data collected by the U.S. Geological Survey (USGS) also demonstrate that cadmium, copper, lead, and zinc are elevated in Silver Bow Creek and Clark Fork River bulk sediments relative to baseline (arsenic was not analyzed in the USGS study). Figure 2-1 (a-d) (data from Lambing et al., 1994) shows that bulk sediments collected from two locations in Silver Bow Creek, at Opportunity and at Warm Springs, contain hazardous substances at concentrations greater than those measured in bulk sediments in Rock Creek (a control stream in the State's Report of Assessment).

Figure 2-2 (a-d) presents the USGS data on hazardous substances in bulk sediments collected from the Clark Fork River (data from Lambing et al., 1994). Bulk sediments were collected from five locations in the Clark Fork River as well as from Rock Creek. Cadmium, copper,

---

<sup>1</sup> Only fine-grained sediment concentrations in control streams were available for comparison in the State's Report of Assessment, but hazardous substance concentrations in bulk sediments of Rock Creek (a control stream) are lower than those in fine-grained sediments (Lambing et al., 1994).



**Figure 2-1. Cadmium (a), Copper (b), Lead (c), and Zinc (d) Concentrations in Silver Bow Creek Bulk Sediments.** Concentrations in control stream (Rock Creek) sediments are shown by the dotted lines. Data are from Lambing et al., 1994.



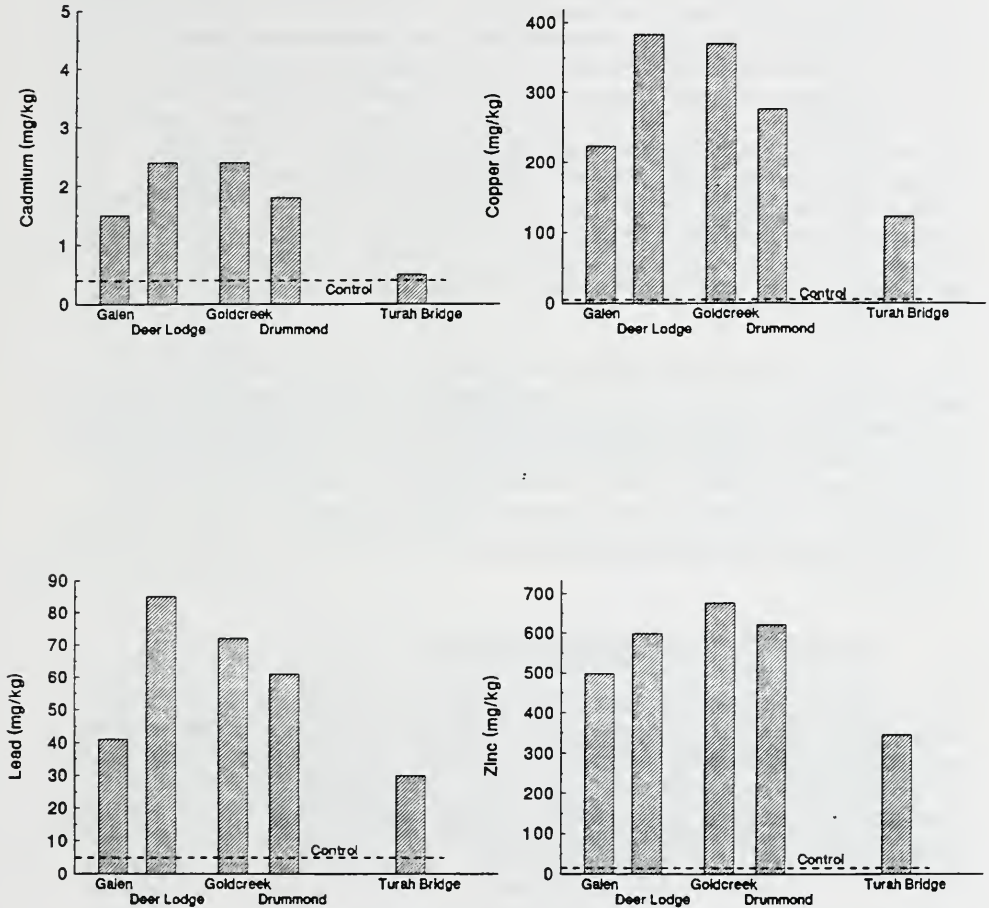


Figure 2-2. Cadmium (a), Copper (b), Lead (c), and Zinc (d) Concentrations in Clark Fork River Bulk Sediments. Sampling stations are ordered from left to right by relative distance from Warm Springs Ponds. Concentrations in control stream (Rock Creek) bulk sediments are shown by the dotted lines. Data are from Lambing et al., 1994.

lead, and zinc concentrations in bulk sediments from the Clark Fork River are all higher than in bulk sediments from Rock Creek. Hazardous substance concentrations measured in Clark Fork River bulk sediments appear to increase downstream of the sampling station near Galen. However, in fine-grained sediments, the USGS and the State (Lipton et al., 1995a) found that hazardous substance concentrations generally decrease downstream of Galen (Lambing et al., 1994). The differences in the observed spatial pattern of hazardous substances between fine-grained and bulk sediments is likely due to varying distributions of sediment grain size in the bulk sediment samples collected. Wilhelm (1986) reports that hazardous substance concentrations differ between grain sizes of Clark Fork River sediments. Since bulk samples do not control for grain size, differences in grain size distribution between samples from different locations confounds a spatial comparison of hazardous substance concentrations. This fact demonstrates the usefulness of controlling for grain size by analyzing only the fine-grained portion.

Table 2-1 presents ratios of hazardous substance concentrations in Clark Fork River fine-grained and bulk sediments with control stream fine-grained and bulk sediments. The ratio of concentrations between Clark Fork River and control stream sediments provides a measure of the relative degree of Clark Fork River sediment contamination compared to control streams. Table 2-1 shows that Clark Fork River bulk sediments are contaminated with hazardous substances to a similar degree as are fine-grained sediments. Therefore, ARCO's contention that fine-grained sediment samples misrepresent ambient conditions is not supported by data collected at the site.

## **2.2    *Natural Mineralization in Sediment Baseline***

ARCO claims that the State's selection of Ruby River, Rock Creek, and Gold Creek as control streams for Silver Bow Creek and Clark Fork River sediment hazardous substance concentrations is inappropriate, because the State fails to take into consideration the fact that in the absence of mining, stream sediments downstream of the Butte ore body "would be expected to be enriched in metals compared to average earth materials" (Ginn, 1995). ARCO claims that baseline metals concentrations for Silver Bow Creek and the Clark Fork River "are not adequately characterized" (Ginn, 1995).

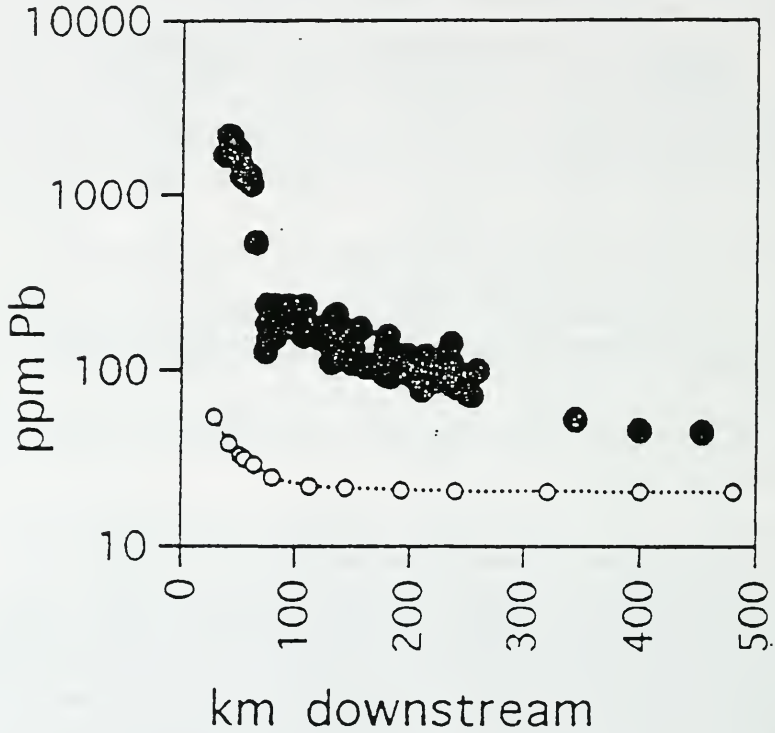
Although studies reported in the geochemical exploration literature do show that stream sediments downstream of natural, unmined ore bodies can contain elevated levels of metals, this fact does not change the State's conclusions regarding the contamination of Silver Bow Creek and Clark Fork River sediments by mining activities. Helgen (1995) developed a model to predict sediment metals concentrations that had been present downstream of ore bodies prior to mining. The model was verified using data from unmined ore bodies. He applied the model to the Silver Bow Creek/Clark Fork River system to predict metals concentrations in sediments prior to mining of the Butte ore body. Figure 2-3 (taken from Helgen, 1995) compares the model's predictions of pre-mining lead sediment concentrations with

**Table 2-1**  
**Ratio of Hazardous Substance Concentrations in Sediments of Clark**  
**Fork River-Upper (headwaters to Garrison Junction) to**  
**Concentrations in Control Stream Sediments**

<b>Hazardous Substance</b>	<b>Ratio in Fine-Grained Sediments<sup>1</sup></b>	<b>Ratio in Bulk Sediments<sup>2</sup></b>
Cadmium	> 26 <sup>3</sup>	> 2.4 <sup>3</sup>
Copper	75	76
Lead	13	13
Zinc	29	34
<sup>1</sup> Data from State's Report of Assessment (Table 3-3). Control streams = Rock Creek, Gold Creek, and Ruby River. <sup>2</sup> Data from Lambing et al., 1994. Control stream = Rock Creek. <sup>3</sup> Ratio is minimum. Cadmium was not detected in control stream sediments.		

concentrations measured in Silver Bow Creek and the Clark Fork River post-mining. The comparison demonstrates that:

- In the absence of mining, sediment metals concentrations in Silver Bow Creek immediately downstream of the ore body are predicted to be elevated relative to concentrations further downstream. However, metal concentrations in Silver Bow Creek are predicted to fall to within 2 times basin-wide background within 15 km downstream of the ore body near Butte. The spatial extent of the predicted elevated concentrations is more than 10 times less than presently occurs (i.e., post-mining), and the predicted elevated concentrations are approximately 10 to 100 times less than presently occur. Thus, a comparison of the conditions predicted in the absence of mining with those that currently occur indicates that mining activities have resulted in large-scale contamination of Silver Bow Creek and the Clark Fork River.
- Throughout Silver Bow Creek and the Clark Fork River, post-mining sediment metals concentrations are much greater than concentrations predicted to occur in the absence of mining. Helgen (1995) concluded that mining activities have amplified the naturally-occurring inputs by 150 times for lead, 530 times for copper, and 1,400 times for zinc.



**Figure 2-3. Sediment Lead Concentrations Predicted in Silver Bow Creek/Clark Fork River in Absence of Mining (open circles) and Recently Measured (solid circles). Figure is from Helgen, 1995.**

Therefore, the State's conclusion that mining activities have greatly contaminated Silver Bow Creek and the Clark Fork River with hazardous substances is not valid.

Additional support for the State's position can be found in the geochemical exploration literature. Several studies demonstrate that sediment metals concentrations are rarely elevated further than 10-20 km downstream of large, unmined ore bodies (Bogoch and Brenner, 1977; Bussey et al., 1993; Bradshaw, 1975, and Lovering and McCarthy, 1978, as cited in Helgen, 1995), including downstream of what at the time was the largest known zinc deposit in Africa (McLaurin, 1978). Similarly, in evaluating possible baseline conditions for Silver Bow Creek and the Clark Fork River, ARCO's consultant, PTI Environmental Services (Deen, 1993), concludes that in climates most similar to Butte, sediment metals concentrations downstream of unmined Cu-Mo porphyry occurrences typically are elevated for 3 to 15 km downstream of the deposit. In contrast, sediments in the Silver Bow Creek/Clark Fork River system are contaminated with metals for at least 220 km downstream of the original ore body near Butte (State's Report of Assessment). Furthermore, as described above, in the uppermost reaches of Silver Bow Creek where metals would be naturally elevated in the absence of mining, metals concentrations post-mining are still approximately 10 to 100 times greater than those expected in the absence of mining.

### 2.3 *Sediments: Conclusions*

ARCO's position is that the State's use of fine-grained sediments to assess contamination in Silver Bow Creek and the Clark Fork River produces biased results and overstates the relative contamination of sediments by metals. An examination of bulk sediment data from Silver Bow Creek and the Clark Fork River demonstrates that:

- Bulk sediments in Silver Bow Creek and the Clark Fork River are highly contaminated with the hazardous substances arsenic, cadmium, copper, lead and zinc relative to baseline.
- The degree of contamination (i.e., concentrations relative to baseline) is similar between bulk sediments and fine-grained sediments of the Clark Fork River.
- The lack of control for grain size in bulk sediment samples confounds spatial evaluation of sediment contamination. Measuring metals concentrations in fine-grained sediments controls for grain size distribution and provides a more accurate means of evaluating spatial trends in sediment contamination.

ARCO also holds that the State's selection of control streams for sediment hazardous substance concentrations is inappropriate, since the selected control streams do not drain areas of natural mineralization, as do Silver Bow Creek and the Clark Fork River. The State's rebuttal demonstrates that:

- ▶ A model of sediment metal concentrations expected in Silver Bow Creek and the Clark Fork River in the absence of mining shows that current sediment contamination is much greater (both in magnitude and spatial extent) than predicted in the absence of mining. Within the first 15 km of Silver Bow Creek, sediment metal concentrations are predicted to fall to within two times basin-wide baseline concentrations. Furthermore, current sediment metal concentrations within the first 15 km are approximately 10 to 100 times greater than the concentrations predicted to occur in the absence of mining.
- ▶ Studies reported in the geochemical exploration literature and a memorandum prepared by PTI Environmental Services, one of ARCO's consultants, show that elevated concentrations of metals typically extend no more than 15-20 km downstream of large unmined ore bodies. In contrast, sediment metals are elevated for at least 220 km downstream of the ore body in Butte.
- ▶ Selected control streams do, in fact, drain areas of natural mineralization, as evidenced by the extensive historic placer mining activity that has occurred on many streams within the State's control drainages (Kerr, Appendix A).



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## CHAPTER 3

### BENTHIC MACROINVERTEBRATES

In the State's Report of Assessment, data on benthic macroinvertebrate communities are used to determine injuries to benthic macroinvertebrates in Silver Bow Creek. Two community metrics, or indicators of invertebrate response to metal contamination, are used: number of taxa; and density of Ephemeropterans. Both of these indices for measuring metals impacts to benthic communities are significantly reduced in Silver Bow Creek relative to control streams.

On September 16-20, 1991, Poulton et al. (1995) collected benthic macroinvertebrates from Silver Bow Creek and a control stream (Rock Creek). Collected invertebrates were enumerated and taxonomically identified. Their results on number of taxa and Ephemeropteran density for Silver Bow Creek and a control stream (Rock Creek) are shown in Table 3-1.

<b>Table 3-1</b> <b>Benthic Macroinvertebrate Community Composition Metrics Silver Bow Creek</b>			
<b>Sample Location</b>	<b>Number of Samples</b>	<b>Mean Number of Taxa (standard deviation)</b>	<b>Mean Ephemeropteran Density (standard deviation)</b>
Silver Bow Creek	4	14.5 (3)*	0 (0)*
Control (Rock Creek)	5	51.6 (1)	8,701 (1,565)
* Significantly lower than control stream mean ( $\alpha = 0.05$ ) using Mann-Whitney test.			
Data from Poulton et al., 1995.			

Table 3-1 shows that the data reported by Poulton et al. (1995) are consistent with those presented in the State's Report of Assessment. Both number of taxa and Ephemeropteran density are significantly lower in Silver Bow Creek relative to the control stream. These data provide additional evidence that demonstrates that the Silver Bow Creek benthic macroinvertebrate community is injured.





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## **CHAPTER 4**

### **SURFACE WATER**

#### ***4.1 Absent Historical and Ongoing Releases of Hazardous Substances, Butte Wastewater Treatment Plant Discharges to Silver Bow Creek would not Discharge Deleterious Levels of Ammonia or BOD***

ARCO asserts that even if hazardous substances were not present in Silver Bow Creek, adverse impacts related to discharge from the Butte Wastewater Treatment Plant (WWTP) would preclude a viable trout population. As described in Kerr (Appendix A), this is not the case; absent hazardous substances, WWTP discharges would be regulated and Silver Bow Creek would support a viable trout population.

#### ***4.2 ARCO Contends that Montana Total Recoverable and EPA Total Recoverable Analytical Methods Yield Similar Results***

Although ARCO has asserted that Montana and EPA Total Recoverable analysis of metals yield similar results, Brand (Appendix G) demonstrates, in fact, that Montana Total Recoverable analyses consistently yield lower values than the EPA Total Recoverable method.



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## CHAPTER 5

### FISHERIES: DETERMINATION OF INJURY

#### 5.1 *ARCO Contends that Failure to Consider the Role of Dissolved Organic Carbon in Toxicity Testing Biased the Results of Testing*

ARCO asserts that dissolved organic carbon (DOC) reduces the toxicity of metals (Ginn, 1995; Jenkins, 1995). Further, ARCO asserts that the State's toxicity testing overestimated lethality and avoidance injuries to trout under ambient conditions by failing to consider the mitigating role of DOC in the Clark Fork River. Three distinct points contradict ARCO's assertion:

- (1) DOC is not present in the Clark Fork River at concentrations typically associated with reduced metal toxicity in the scientific literature.
  - (2) Mitigating effects of Clark Fork River DOC likely are less than those identified in the literature cited by ARCO. Moreover, the effect of DOC on metal toxicity has been found to delay lethality rather than reducing it. Short term testing of the effects of DOC may have attributed toxicity reductions that were not present.
  - (3) DOC does not reduce behavioral avoidance.
- 1. DOC is not present in the Clark Fork River at concentrations associated with reduced metal toxicity.**

Several authors have found that DOC can reduce the toxicity of copper and other hazardous metals. However, concentrations of DOC shown to cause this reduced toxicity are typically substantially higher than are found in the Clark Fork River. This is supported by the literature cited by ARCO's expert report (Jenkins, 1995); the empirical data from these studies demonstrate that elevated DOC concentrations of both synthetic and natural sources can mitigate metal toxicity in some cases, but only at DOC concentrations higher than those reported for the Clark Fork River. Furthermore, as discussed below, it is the complexation characteristics (number of binding sites, or "capacity," and binding strength, or "affinity") of DOC for particular metals, as well as relative concentrations of the DOC and metal, that determine whether DOC will mitigate metal toxicity.

Jenkins (1995) cites several studies to claim that DOC is particularly important in controlling the speciation, bioavailability and toxicity of copper in most freshwater systems. However,

these citations do not support ARCO's conclusion that Clark Fork River DOC causes toxicity reductions. For example:

- (1) Sunda and Lewis (1978) found that DOC from the Newport River (which contained approximately 22 mg/l DOC) mitigated copper toxicity in algae with dilution concentrations of 7 and 20 mg/l DOC. In other words, this study demonstrated toxicity reductions, to algae, at DOC concentrations several-fold greater than those in the Clark Fork River which typically has DOC concentrations in the range of 1-4 mg/L (see below)..
- (2) Sunda and Hanson (1979) empirically derived conditional stability constants of organic and inorganic ligands for copper using titrimetric methods in order to investigate copper complexation characteristics of two rivers in coastal North Carolina. Although referenced by ARCO, this citation does not address the effects of DOC on metal toxicity.
- (3) Zamuda et al. (1985), testing american oysters in salt water, found that copper accumulation rates were reduced in the presence of DOC derived from diatoms. However, the relevance of this study to the Clark Fork River -- in which toxicity was not evaluated, the DOC was derived from diatoms, and testing was performed in salt water with oysters -- is unknown. Moreover, the authors found that the effects of DOC were greatly reduced by ultraviolet light. This suggests that the effects of DOC in nature may be even less than in laboratory experiments cited by ARCO.
- (4) Giesy et al. (1986) empirically determined complexation parameters for copper by humic acid (DOC) fractions from surface waters in the southeastern U.S.. Again, this citation provided no direct data regarding toxicity or bioavailability issues addressed.
- (5) Cabaniss and Schuman (1988) empirically derived conditional stability constants and other parameters using titrimetric methods in an effort to describe copper binding with Suwannee River fulvic acid (DOC); no toxicity or bioavailability issues were addressed.
- (6) Playle et al. (1993a, 1993b) evaluated the protective effects of both natural and synthetic DOC sources on copper and cadmium toxicity to fish. These studies report that below 4.8 mg/l DOC, little or no protective effect of natural DOC source was observed. However, DOC concentrations  $\geq 4.8$  mg/l decreased copper binding to gills.
- (7) Welsh et al. (1993) developed an empirically derived model that showed both natural DOC over the range of 0.2-16 mg/l and pH over the range of 5.4-7.3 explained 93% of the variability in change in 96-h LC<sub>50</sub> estimates for fathead minnows. However in the presence of DOC concentrations less than 5 mg/l, it was higher pH, not higher DOC concentrations, that caused the observed reductions in copper toxicity. Therefore,

the data from Welsh et al. (1993) further support the other studies showing natural DOC concentrations  $\leq 5$  mg/l do not alter copper toxicity.

Further, in laboratory tests performed by Hagler Bailly Consulting and the University of Wyoming (Marr et al., 1995), concentrations of organic acids simulating 4 mg/L of ambient DOC in an Idaho stream did not reduce the toxicity of copper to rainbow trout. Concentrations of organic acids simulating as high as 8 mg/L of ambient DOC caused negligible toxicity reductions.

Ambient DOC concentrations in the Clark Fork River typically are substantially lower than those DOC concentrations found to reduce toxicity in the above studies. DOC concentrations measured in the Clark Fork River over the period of May 1992 - August 1994 ranged from 1-4 mg/L DOC (Table A-14, Appendix A, Lipton et al., 1995a), with concentrations typically near 2 mg/L DOC. This concentration that has not been associated with reduced metal toxicity. These data contradict ARCO's assertion that the State's testing overestimated toxicity.

**2. It is the composition, rather than the concentration, of ambient DOC that causes toxicity reduction.**

As described above, ARCO's assertion that toxicity in the Clark Fork River is reduced by the presence of DOC is based on literature in which DOC concentrations were higher than those in the Clark Fork River. In addition, ARCO's assertion assumes that all DOC acts similarly in reducing metal toxicity. However, in controlled testing performed by Hagler Bailly Consulting and the University of Wyoming (Marr et al., 1995), we found that this is not the case. The potentially mitigating effects of DOC on metal toxicity were found to be dependent on the metal binding affinity and capacity of the DOC. In our testing, we found that organic acids with metal binding affinities less than those of a trout gill did not reduce toxicity (in other words, the trout gill essentially out-competes the organic acid for the metal). A substantial portion of the DOC present in Panther Creek, a stream located in Idaho southwest of Missoula, MT, was present as organic compounds that did not reduce copper toxicity to rainbow trout (Marr et al., 1995). Much of the laboratory testing on the effects of DOC in reducing metal toxicity has been performed using commercially developed humic acids, synthetic organic chelators, or DOC derived from microcosm media [e.g., Aldrich humic acid, EDTA (ethylenedinitrilotetraacetic acid), NTA (nitrilotriacetic acid); see Playle et al. 1993a, 1993b; Meador, 1991; Winner and Gauss, 1986; Pärt and Wikmark, 1984; Shaw and Brown, 1984; Anderson and Morel, 1978]. Our testing indicated that the "toxicity-mitigating" properties of these materials are substantially greater than ambient DOC in a Rocky Mountain stream (Marr et al., 1995). Interestingly, the Zamuda et al. (1985) reference cited by ARCO also indicates that "natural dissolved organics differ markedly with respect to their influence upon copper bioavailability."

Jenkins (1995) also claims that copper-binding sites on natural DOC exceed the affinities for copper-binding sites on fish gills, based again on a misleading interpretation of Playle et al., (1993a, 1993b). ARCO's assertion is based on a model in Playle et al. (1993a). This model was derived from data from experiments using synthetic DOC sources (EDTA, NTA, EN, citrate, oxalate, and glutamate, and salicylate). Upon testing the model, the modeled gill-copper data *underpredicted* the measured gill-copper concentrations in all but one of the actual lake waters tested. In other words, the model overestimated toxicity reductions. Moreover, the measured data in these studies showed that DOC simulating 5 mg/l of DOC failed to reduce copper or cadmium accumulation on fish gills, and it did *not* alter metal toxicity.

Further, in bioassays simulating surface waters for a site in Idaho, we found that DOC can delay mortality without reducing the absolute level of mortality (Marr et al., 1995). For example, the median survival time for rainbow trout exposed to 20 µg/l copper with 4 or 8 mg/L DOC was nearly twice that of the 0 mg/L DOC treatment. However, the absolute mortality level after 10 days of exposure was not different between the three DOC treatment levels, with about 90% mortality in all DOC treatments at 20 µg/l copper. These data suggest that short-term experiments (e.g., < 96-hours) may substantially overestimate toxicity reductions attributable to DOC (see comments on Water Effects Ratio testing, below).

Therefore, ARCO's reliance on the literature to claim that DOC in the Clark Fork River reduces metal toxicity overestimates the importance of DOC on toxicity reductions by (1) failing to account for the fact that most literature reports rely on testing with DOC substances (e.g., EDTA, NTA) that likely have a much greater ability to reduce toxicity than conditions in the Clark Fork River, and (2) inferring DOC effects from short-term tests in which toxicity "reductions" attributable to DOC may actually have been situations in which mortality was delayed, not reduced.

### **3. DOC does not reduce avoidance behavior by salmonids.**

ARCO asserts that the State's avoidance testing overestimated behavioral avoidance injuries by failing to consider the role of DOC in reducing avoidance responses. We are aware of no scientific study that supports this assertion, nor is any provided by ARCO. To the contrary, in controlled laboratory testing performed by Hagler Bailly Consulting and the University of Wyoming over a range of DOC concentrations (0-8 mg/L DOC) and a range of pH (6.5-8.5) we found that DOC had no discernable effect on the avoidance of copper by rainbow trout and chinook salmon (Marr et al., 1995). These data directly contradict ARCO's assertions, which themselves are not substantiated with data or literature.



## **5.2    *Water-Effects Ratio (WER) Testing Performed by ARCO was Flawed***

ARCO relies on water-effects ratio (WER) testing performed by Stubblefield (1995) to suggest that the toxicity of copper in ambient Clark Fork River water is less than that in “clean” laboratory waters. The tests consisted of short-term (typically 48-96 hour) (note above comments documenting that short-term testing may underestimate toxicity) bioassays using ambient Clark Fork River water and reconstituted laboratory water (Stubblefield, 1995). The reliance on WER testing and toxicity testing methods performed for ARCO is flawed for the following reasons:

- (1)    WER testing performed for ARCO was inappropriate for evaluating copper toxicity in the Clark Fork River.
- (2)    WER testing performed for ARCO ignored primary contaminant sources and exposure pathways in the Clark Fork River.
- (3)    WER testing performed for ARCO failed to follow specified U.S. EPA and ASTM (American Society for Testing and Materials) guidelines for conducting toxicity tests.

### **1.    *WER testing performed for ARCO was inappropriate for evaluating copper toxicity in the Clark Fork River.***

The WER procedure, as defined by the U.S. EPA guidance (U.S. EPA, 1994), is inappropriate if the effluent, upstream water, and or downstream water are toxic to the test organism. Surface waters in the Clark Fork River have previously been demonstrated to contain metals concentrations that are toxic to various species of aquatic organisms, including rainbow trout, as evidenced by recent fish kills and *in situ* bioassays (Lipton et al., 1995a); background acute toxicity was also observed during the WER testing performed for ARCO (Stubblefield, 1995). Therefore, conducting a WER testing program to evaluate copper toxicity is inappropriate. Furthermore, because existing data demonstrate that multiple metals are toxic to biota in the Clark Fork River and because the WER testing program performed for ARCO only accounts for a single metal (i.e., copper only), the data collected during the WER testing program are inadequate for evaluating the multiple-metal situation that exists and influences ambient surface water toxicity in the Clark Fork River.

In addition, the WER testing program performed for ARCO was inappropriate for evaluating temporal and spatial changes in the toxicity of Clark Fork River surface waters because site water sampling and experimental designs failed to account for the extreme of water conditions that occur in the river basin, including upstream and downstream waters. As described in the U.S. EPA WER guidance document (U.S. EPA, 1994), the experimental design should test certain assumptions about variations in endpoint parameters with respect to time, location, and depth within the site. To adequately test variations in the WER endpoints used for adjusting copper criteria due to site specific conditions in the Clark Fork River it would be necessary to

conduct toxicity tests using paired site and laboratory waters over the full range of metals concentrations (in addition to copper) and over the range of hardness and alkalinity conditions that occur.

Additionally, the U.S. EPA recommends an experimental design for a WER testing program to emphasize intensive sampling and testing during the more extreme seasons known to influence discharge events (U.S. EPA, 1994). For the Clark Fork River, the most extreme site conditions that influence metal releases into surface waters occur during spring high flows and during thunderstorms. The WER testing program conducted for ARCO emphasized sampling events during low flow conditions (i.e., 7 of the 9 sampling events, >75%, for all of Phase I, II, and III; Stubblefield, 1995). Thus, the experimental design of ARCO's WER testing program was inappropriate for evaluating site specific conditions that influence copper toxicity in the Clark Fork River.

## **2. WER testing performed for ARCO ignored primary contaminant sources and exposure pathways in the Clark Fork River.**

In the Clark Fork River, a primary source of contamination is the bed sediments (Lipton et al., 1995a). Data collected by Essig and Moore (as cited in Lipton et al., 1995a) show that bed sediments throughout Silver Bow Creek and the Clark Fork River from Warm Springs Ponds to Milltown Reservoir contain elevated concentrations of hazardous substances. Furthermore, food sources for trout (e.g., benthic macroinvertebrates) accumulate metals in the Clark Fork River and provide a critical pathway by which trout and other aquatic biota are exposed to metal contaminants (Lipton et al., 1995a). Similarly, studies performed for ARCO demonstrated that trout suffer growth reduction injuries when they consume contaminated zooplankton (see section 5.4) which, in turn, are exposed to hazardous substances in the water column. The WER testing program conducted for ARCO ignored both the bed sediments as a source of toxicity and contaminated invertebrates as a pathway for food-chain exposure to trout. Therefore, ARCO's WER testing does not adequately address deleterious exposures of hazardous substances to trout.

While sediment criteria remain unavailable from the U.S. EPA for deriving water quality criteria, it is the U.S. EPA's contention that total recoverable metals (i.e., particulate metals) can contribute to the overall toxicity to aquatic ecosystems in locations where the water column is not the only route of exposure. The State of Montana's adoption of a total recoverable method for assessing ambient water quality criteria in the Clark Fork River is consistent with the U.S. EPA's guidance for considering conservative approaches in evaluating water quality criteria, especially in situations where sediments and food-chains are important pathways of metal exposures (Prothro, 1993)(Bergman, Appendix I).



### **3. WER testing performed for ARCO failed to follow specified U.S. EPA and ASTM guidelines for conducting toxicity tests.**

Many of the testing method guidelines that are required by the U.S. EPA in conducting WER testing programs and required by ASTM for conducting toxicity tests were apparently not followed in the toxicity tests conducted for ARCO's WER testing program. In addition to not meeting specified criteria and guidances provided by U.S. EPA and ASTM, the WER testing programs conducted for ARCO failed to use scientifically defensible approaches for toxicity testing in certain testing procedures. As a result, the data obtained from ARCO's WER testing program should not be used because of test criteria violations and nonadherences to specified guidances. Certain flaws in the toxicity testing procedures performed for ARCO are described below.

#### *Nonrenewal of Test Solutions*

A majority of the toxicity tests performed in ARCO's WER testing program utilized static conditions when renewal of test solution is required according to U.S. EPA specified guidelines for conducting WER testing (U.S. EPA, 1994). As stated in the WER guidance (U.S. EPA, 1994), "tests whose duration is longer than 48 hours must be renewal tests." This requirement was not met in the Phase I/II studies conducted for ARCO (Stubblefield, 1995), where acute toxicity tests with rainbow trout and fathead minnows consisted of static test conditions and test solutions were not renewed throughout the 96 hours of testing (ENSR Project No. 8505-093-130, Protocol No.s OM2MT.130.10A and PPM2MT.130.11A, April 1993). (ARCO did not provide protocols for determining test conditions in Phase III studies. This report may be supplemented following receipt and review of the protocols.)

Moreover, ARCO is relying on the rainbow trout 96-hour acute toxicity test as the "Primary" test for determination of a FWER (Stubblefield, 1995), which places primary importance on the test conditions used during the rainbow trout test. The rainbow trout as a test organism has biological and physiological requirements that do not make them acceptable for static testing, including their: size, respiratory and feeding requirements, temperature tolerance, growth rate, and dissolved oxygen sensitivity. For example, the U.S. EPA's national AWQC for copper was based on bioassays conducted with four salmonid species, fathead and bluntnose minnows, and bluegill. The majority of tests conducted since 1970 have used flow-through toxicity testing methods (U.S. EPA, 1984). Of the 51 referenced bioassays with trout (rainbow trout, cutthroat trout, and brook trout) used by the U.S. EPA for establishing national AWQC for copper, only one of those 51 bioassays was conducted with static or static-renewal testing methods (U.S. EPA, 1984). This bioassay reported an LC<sub>50</sub> value of 890 µg/l copper — nearly 20 times greater than the promulgated acute AWQC value for the hardness at which the test was conducted (hardness of 290 ppm), and many times greater than LC<sub>50</sub> values quantified using flow-through testing. For comparison, the LC<sub>50</sub> estimates obtained during the WER testing program for rainbow trout in site waters ranged from 86 to 600 µg/l total recoverable copper (Stubblefield, 1995). Again, these ARCO values are elevated

relative to those references cited by U.S. EPA (1984) in its water quality criterion document. Had ARCO's WER testing program been conducted with flow through (or even static renewal) testing conditions specified by U.S. EPA and ASTM, the resulting endpoints (i.e., LC<sub>50</sub> values) used to determine WER would likely have been substantially lower.

#### *Match of Laboratory Waters to Site Waters*

Alkalinity and hardness are to be equivalent between the site and laboratory waters for WER testing programs (U.S. EPA, 1994); according to the ENSR protocols this was a primary objective in the test methods. However, the reported chemical characterizations of site and laboratory waters showed that in all cases hardnesses and/or alkalinities in laboratory waters were less than that of site waters (Phase I and II studies; ENSR Consulting and Engineering, 1995) (Table 5-1). The table demonstrates a clear bias in the ARCO testing: in 14 of the 15 hardness comparisons and 15 of 15 comparisons for alkalinity, site water had lower values than laboratory waters. Further, chemical characterizations of the site and laboratory waters from these same WER studies showed lower pH values in laboratory waters compared to site waters in a majority of the cases (Phase I and II studies; ENSR Consulting and Engineering, 1995). Having lower hardness, alkalinity, and/or pH in laboratory waters will cause an artificial difference between the test endpoints for site waters and laboratory waters, such that lower LC<sub>50</sub> values will be obtained for laboratory waters compared to site waters. Thus, the WERs obtained during Phase I and II studies will misrepresent any differences between site and laboratory waters because the two waters were not sufficiently matched for hardness and alkalinity as required by the U.S. EPA guidance (U.S. EPA, 1994). The result of this mismatch will be to inappropriately inflate WER values.

#### *Inadequate Replication and Numbers of Test Organisms*

Inadequate replication of exposure treatments during the WER testing program was used in the acute toxicity tests using rainbow trout. According to the ENSR project protocols (ENSR Project No. 8505-093-130, Protocol No. OM2MT.130.10A, April 1993) only two replicates were tested per treatment. A higher level of replication should have been used, as was done with testing for fathead minnows and ceriodaphnia (i.e., four or ten replicates), especially given the relative importance placed on the rainbow trout as the primary species of concern for the WER testing program.

In addition, inadequate numbers of test organisms in exposure replicates were used in the acute toxicity tests with fathead minnows. According to the ENSR project protocols (ENSR Project No. 8505-092-130, Protocol No. PP2MT.130.11A, April 1993) only five fathead minnow individuals were tested per replicate. This low level of replication decreases the power of the testing.

**Table 5-1**  
**Comparison of Site and Laboratory Water Hardness (Alkalinity) Used in ARCO**  
**Water Effects Ratio (WER) Phase I and II Testing:**  
**When “Site” Water is Greater than “Lab” Water, WER is Biased Upwards**  
**(data from ENSR Consulting and Engineering, 1995)**

Location	Site Water	Laboratory Water	Lab or Site Greater?
<b>Warm Springs</b>			
Test 1	164 (95)	156 (85)	Site (Site)
Test 2	118 (70)	110 (61)	Site (Site)
Test 3	124 (82)	118 (76)	Site (Site)
Test 4	134 (89)	130 (78)	Site (Site)
Test 5	158 (69)	148 (64)	Site (Site)
<b>Clark Fork River 1</b>			
Test 1	264 (110)	252 (100)	Site (Site)
Test 2	200 (92)	170 (82)	Site (Site)
Test 3	116 (74)	110 (71)	Site (Site)
Test 4	198-200 (114-115)	180 (113)	Site (Site)
Test 5	262 (100)	264 (95)	Lab (Site)
<b>Clark Fork River 2</b>			
Test 1	250 (139)	224-216 (125-118)	Site (Site)
Test 2	248 (140)	244 (131)	Site (Site)
Test 3	146 (102)	138 (95)	Site (Site)
Test 4	200-202 (136-138)	180 (131)	Site (Site)
Test 5	244 (158)	230 (145)	Site (Site)

### *Feeding During Acute Toxicity Testing*

According to the test guidelines used (U.S. EPA, 1994; ASTM, 1991), test organisms should not be fed during acute toxicity testing. However, according to the project protocols for the WER testing program, test organisms were fed a concentrated suspension of newly hatched brine shrimp *ad libitum* during the conduct of the acute toxicity tests with fathead minnows (ENSR Project No. 8505-092-130, Protocol No. PP2MT.130.11A, April, 1993). Feeding the test organisms during acute toxicity testing will very likely alter the toxicity thresholds produced by modifying the toxicity of copper in solution and by compromising the physiological state of the test organisms (ASTM, 1991). The effect of this will be to underestimate toxicity.

### *Duration of Exposure*

The acute toxicity tests performed for ARCO's WER testing program used 48-96 hour exposure durations under static test conditions. However, according to the ASTM guidelines (ASTM, 1991), it is recommended that acute toxicity tests conducted for determining LC<sub>50</sub> values as endpoints include an extended exposure duration of 96 hours, for a total exposure duration of 192 hours. This would allow determining whether additional organisms are affected or killed after the initial 96 hours (ASTM, 1991), and subsequently prevent an underestimation of the toxicity of the test solution.

Moreover, Marr et al., (1995) showed that short-term testing (e.g., 48-h) underestimated toxicity that occurred over a longer observation period. Thus, the short-term tests performed by ARCO may have similarly underestimated toxicity of site water that might have otherwise been observed over a longer duration test.

### *Acceptability of Tests*

According to the criteria for test acceptability specified by the ASTM guidance (ASTM, 1991), there are multiple criteria that must be met for determining the acceptability of the test. The following list describes certain ASTM criteria that should be evaluated for determining the acceptability of the toxicity studies conducted for ARCO's WER testing program in the Clark Fork River.

1. According to ASTM a toxicity test would not be acceptable if "any individual measured temperature in any test chamber was more than 3°C different from the mean of the time-weighted average measured temperatures for the individual test chambers." This criteria was likely violated; on at least one laboratory audit it was discovered that temperature fluctuations during at least one day for the rainbow trout waterbath fluctuated by 6°C (temperature rose from 12°C to 18°C) (ENSR Consulting and Engineering, Memorandum, October 27, 1993).

2. According to ASTM a toxicity test would not be acceptable if "the test organisms were not maintained in the dilution water at the test temperature for at least the last 48 h before they were placed in test chambers." This criteria was likely violated; "rainbow trout will not be acclimated in dilution water for 48 hours prior to testing. Rather, trout will be acclimated to test temperature in moderately hard laboratory reconstituted water (hardness 80-100 mg/L as  $\text{CaCO}_3$ , alkalinity 60-70 mg/L as  $\text{CaCO}_3$ ). This change was approved by Tom Willingham prior to the initiation of Phase I testing" (ENSR Consulting and Engineering, Letter to ARCO, April 15, 1993).
3. According to ASTM a toxicity test would not be acceptable if "treatments were not randomly assigned to individual test chamber locations." It is unclear from the project protocols whether this criterion was met or not (ENSR Consulting and Engineering, Memorandum, January 13, 1993).

### **5.3 *Food-chain Testing Performed by ARCO Demonstrates that Exposure to Hazardous Metals in Diets causes Growth Reduction Injuries in Trout***

ARCO has asserted that laboratory testing performed by ENSR Consulting and Engineering (ENSR) under contract to ARCO demonstrates that food-chain exposure to metals does not cause growth reduction injuries to trout (see Stubblefield, 1995). This conclusion is based on tests performed using unnatural diets (brine shrimp) (Mount et al., 1994) and invertebrates (primarily *Daphnia*) collected from the Warm Springs Ponds (ENSR, 1995).

As described in greater detail by Woodward et al. (Appendix B), the Mount et al. (1994) study is not appropriate to evaluating food-chain effects in the Clark Fork River. Further, independent statistical analysis of the ENSR (1995) data by Woodward et al. (Appendix B) demonstrate clear growth reduction injuries from dietary exposure to contaminated zooplankton from the Clark Fork. Thus, ARCO's own data demonstrate that food-chain exposure to metals-contaminated invertebrates from the Clark Fork River causes growth reduction injuries.

### **5.4 *Observed Reductions in Trout Populations are not Attributable to Placer Mining in the Clark Fork River***

ARCO contends that trout population reductions observed in Silver Bow Creek/Clark Fork River may be attributable to placer mining in Silver Bow Creek and Clark Fork River.

As shown by Kerr (Appendix A), any potential effects of placer mining are accounted for by the state's fisheries population sampling.



**5.5 *ARCO Asserts that Hardness Concentrations used in the State's Toxicity Tests were not Representative of those Found in the Clark Fork River***

ARCO has asserted that the 100 mg/L hardness concentration employed in the toxicity tests performed for the State is substantially less than ambient hardness concentrations in the Clark Fork River. It is emphasized that test conditions employed in the fish toxicology studies were intended to represent spring conditions in the Clark Fork River when hardness concentrations are generally at their lowest levels and metals concentrations tend to be at their highest concentrations. This combination of low hardness and high metals concentrations are potentially the most toxic conditions during the year and therefore represent a critical limiting factor for trout populations.

Kerr (Appendix A) analyzed hardness data from three sites on the Clark Fork River (Deer Lodge, Gold Creek and Turah) for the years 1989 to 1994. Based on this analysis, the hardness concentrations utilized in toxicity testing performed for the State is appropriate to simulating critical spring conditions.

**5.6 *Additional Behavioral Avoidance Testing Performed by the State Confirms that Trout Prefer Uncontaminated Water Typical of Tributaries to Contaminated Clark Fork River Water. Testing also Confirmed that Copper and Zinc are the Cause of Behavioral Avoidance Injuries to Trout***

In the report of Jenkins (1995), ARCO contends that behavioral avoidance injuries documented by the State were "unrealistic" because the control water used in the avoidance challenges did not contain metals and "dissolved metals are present in the surface waters of the tributary streams which feed the Clark Fork River . . ." Additional testing performed by the State (Woodward and Bergman, Appendix D) negates this argument by showing that:

- trout, including endemic species, avoid contaminated Clark Fork River water relative to cleaner tributary waters. Thus, trout suffer behavioral avoidance injuries in Silver Bow Creek/Clark Fork River.
- behavioral avoidance injuries are caused by exposure to the hazardous substances copper and zinc.

**5.7 *Total Recoverable Metals Criteria are More Appropriate than Dissolved Criteria for the Clark Fork River System***

The State of Montana currently regulates hazardous metals in the Clark Fork River based on total recoverable metals. ARCO has argued that dissolved metals are more appropriate to evaluating and regulating the adverse effects of metals on aquatic biota. As described in

Bergman (Appendix H), use of EPA total recoverable metals would be more appropriate to regulate hazardous metals in the Clark Fork River because of (1) heavy contamination of bed sediments with hazardous metals, (2) injurious food-chain exposures to hazardous metals, and (3) temporal and spatial variability in ambient water quality conditions in the Clark Fork River.

**5.8 *It is Inappropriate to Compare the Results of Food-chain Studies Performed by ARCO using Artificial Laboratory Diets and Food-chain Studies Performed for the State using Natural Diets from the Clark Fork River***

As shown in Woodward and Bergman (Appendix I), biochemical differences exist between metals bound to brine shrimp (as were used in dietary exposure studies performed for ARCO) and the natural Clark Fork River invertebrate diets used in the food chain testing performed for the State. For example, differences existed in (1) dietary partitioning of hazardous substances from the diets, (2) digestion of the diets, and (3) concentrations of essential amino acids. Therefore, it is inappropriate to compare the results of food-chain studies performed by ARCO using artificial laboratory diets and food-chain studies performed for the State using natural diets from the Clark Fork River.





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## CHAPTER 6

### FISHERIES: QUANTIFICATION OF INJURY

ARCO criticized the State's quantification of injury to trout populations on several grounds. This Chapter addresses key aspects of these ARCO opinions.

#### **6.1    *ARCO Contends that the State did not Consider the Effects of Channelization in Quantifying Differences between Trout Populations in the Clark Fork River and in Reference Sites***

ARCO contends that the State failed to consider the effects of channelization on trout populations in the Clark Fork River (Reiser, 1995 and Wesche, 1995). Further, ARCO indicates that channelization in the Clark Fork River is substantially greater than at reference sites utilized by the State (see Lipton et al., 1995a, Chapter 6), and that this channelization is the cause of differences in trout populations observed between the Clark Fork River and reference sites. ARCO's argument assumes that (1) the Clark Fork River and Silver Bow Creek have a greater degree of channelization than reference sites with resulting changes in stream morphology and fish habitat, (2) the State's site selection methodology failed to account for potential habitat differences caused by channelization.

#### **1.    Channelization in the Clark Fork River/Silver Bow Creek is similar to reference sites.**

Reiser (1995) stated that about 50 miles (or, according to Reiser, about 50%) of the Clark Fork River have been channelized. Wesche (1995) estimated that approximately 60 miles (according to Wesche, about 48%) of the Clark Fork River have been altered. Jensen (Appendix G), however, determined that approximately 33% of the Clark Fork River (or about 40 miles) have some degree of alteration. Chapman (Appendix D) presents data showing that the degree of channelization (about 33%) in the Clark Fork River is similar to reference streams. For example, Chapman (Appendix D) cites data from Peters and Alvord (1963) showing that approximately 30% of the Bighole River has been channelized. Similarly, Chapman (Appendix D) indicates that channelization has occurred in the Ruby River and Flint Creek, reference streams used by the State.

Moreover, as described by Chapman in Appendix D, channel modifications can vary substantially in the degree to which stream morphology is affected. Although the overall floodplain of the Clark Fork River is extensively modified, the active channel is not constrained to the degree suggested by ARCO. Chapman (Appendix D) points out that

ARCO's data show stream sinuosity (a measure of the degree to which the active channel of a stream or river meanders) in all reaches in the Clark Fork River is equal to or greater than the Big Hole River and Rock Creek. Similarly, sinuosity in the Clark Fork River (from Gold Creek to Flint Creek) exceeded that of the reference streams Flint Creek, Rock Creek, and the Big Hole River. These data suggest that, if anything, the effects of channelization on stream morphology were smaller in the Clark Fork River than in matched reference areas.

**2. Channelization in the Clark Fork River/Silver Bow Creek has not degraded habitat relative to reference sites.**

Chapman (Appendix D) analyzed differences in trout habitat in fish sampling sites in channelized sites in the Clark Fork River and "straight" sites in reference streams. As shown in Table 6-1, Chapman (Appendix D) found that in some 80% of the cases in which instream microhabitat differed between the Clark Fork River and reference sites, the difference favored the Clark Fork River. Again, this shows that, if anything, the Clark Fork River should have *greater* trout populations than the reference sites.

Further, Chapman (Appendix D) compared weighted usable area (WUA) — another indicator of available habitat — between six channelized sites in the Clark Fork River and matched straight reference sites. This comparison demonstrated that the Clark Fork River contained **more** habitat for brown trout adults in 5 of 6 cases (83%). Again, these differences suggest that the Clark Fork River should contain *more* trout than reference sites. In 4 of 6 cases (67%), the reference sites contained more habitat for rainbow trout adults than the Clark Fork River. However, Chapman points out that in 2 of these 4 cases, the magnitude of difference in WUA is small (approximately 10%). Therefore, for rainbow trout, any differences in WUA were relatively minor — particularly when viewed in the context of the several fold reduction in rainbow trout populations in the Clark Fork relative to reference streams.

Finally, Appendix D addresses ARCO's contention that the measurement of trout populations in "altered" stream segments in the Clark Fork River has exerted a downward bias on trout population estimates. Chapman demonstrates that inclusion of altered reaches in the quantification of trout population differences between the Clark Fork River and reference sites does not artificially inflate the observed population reductions. Indeed, Chapman found that inclusion of the altered sites diminished the magnitude of population change.<sup>1</sup> This observation is inconsistent with the conclusion that the State's population estimates were biased by sampling altered reaches.

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<sup>1</sup> Chapman (Appendix D) demonstrates that the mean reduction in adult brown trout populations between the Clark Fork River and reference streams is approximately seven-fold when altered sites are included in the analysis and approximately thirteen-fold when altered sites are eliminated from the analysis.

**Table 6-1**  
**Comparison of Microhabitat Differences: Clark Fork River and Reference Sites**  
**(from Chapman, Appendix D)**

Test Reach	# Habitat Features Significantly Different	# Habitat Features Not Significantly Different	% of Habitat Features Where Significant Differences Favor Clark Fork River
State 4 Reach 1	5	5	100%
State 4 Reach 2	3	7	66%
State 4 Reach 3	2	8	100%
State 4 Reach 4	1	9	100%
State 4 Reach 5	5	5	80%
State 4 Reach 6	4	6	50%
<b>Total</b>	<b>20</b>	<b>40</b>	<b>80%</b>

Thus, ARCO's contention that channelization in the Clark Fork River was not considered in the State's sampling methodology is not valid: channelization in the impact and control rivers was generally similar, effects on stream morphology (as evidenced by sinuosity data) were not apparent between the Clark Fork River and reference sites (or favored the Clark Fork River), microhabitat data generally indicated that habitat in the Clark Fork River was greater in the Clark Fork River than in reference sites, and macrohabitat data (as incorporated by WUA) was generally greater in the Clark Fork River.

## **6.2 ARCO Contends that Elevated Water Temperatures in the Clark Fork River Cause Trout Population Declines Relative to Reference Streams**

Differences in water temperatures occur between the Clark Fork River and reference sites (see Lipton et al., 1995a, Appendix A). ARCO contends that these differences account for differences in trout populations (Reiser, 1995). However, Chapman (Appendix D) concludes

that such differences in water temperature are not biologically significant and that differences in trout populations are not attributable to water temperatures.

As a related matter, ARCO's assertions regarding elevated temperatures in the Clark Fork River relate to agricultural dewatering of the Clark Fork (with resultant decreases in flows and increases in temperatures during summer months). Such land use alterations occur in the State's reference rivers, as well. For example, the Big Hole River, which served as a reference river for several Clark Fork River reaches, was recently determined to meet the criteria for designation as a "chronically dewatered" stream by the Montana Department of Natural Resources and Conservation (MDNRC, 1995). MDNRC (1995) concludes that dewatering, caused by agriculture and other land uses, has caused elevated stream temperatures, increased algae, reduced dissolved oxygen, and limits trout fisheries in the Big Hole River. Further, MDNRC (1995) indicates that stream temperatures considered lethal to fish have been recorded in the Big Hole. Therefore, the Big Hole River, and other reference sites used by the State, are subject to similar land uses as the Clark Fork, and accurately represent baseline conditions.

### **6.3    *ARCO Asserts that the State's Fish Population Sampling was Biased because of Methodological Deficiencies***

Reiser (1995) suggests that the State's fish population sampling was biased because (1) counts were made during the day, and (2) the State performed multiple-pass snorkeling in wide stream segments. Hillman and Chapman (Appendix E) report the results of additional snorkeling studies performed to evaluate ARCO's critique. As a result of these studies, Hillman and Chapman conclude that nighttime snorkeling counts are less reliable than daytime counts performed by the State, and that multiple-pass and single-pass surveys do not bias fish population counts. These conclusions contradict ARCO's assertion that the State's snorkeling methodology biased trout population estimates.

### **6.4    *ARCO Asserts that the Clark Fork River and Reference Sites Differ in Terms of Land Use***

ARCO has criticized the State's hierarchical classification procedure, claiming that the stratification methodology did not account for land use distinctions across drainage basins. In his opinion, Jensen (Appendix F) documents that the Clark Fork River and reference stream basins contain similar types and frequencies of land uses: urban uses, agricultural uses, rangeland, forest, open water, wetland, barren lands, and tundra (see Appendix F, pp. 15-16). Therefore, ARCO's critique that land uses are not accounted for in the classification process employed by the State is not substantiated by actual land use data.

### 6.5 Trout Population Reductions: Evaluation of Causality

A number of environmental and anthropogenic factors can alter trout populations. The State has determined that trout populations have been reduced as a result of hazardous substance releases. However, ARCO has asserted that the significant causative factors are: channelization, human land uses, excess nutrients, elevated temperatures, and reduced dissolved oxygen. Table 6-2 presents an analysis of the *consistency* of each of these factors with data relevant to the State's injury assessment.

As shown in the table, only hazardous substances are consistent with observed conditions in the Clark Fork River/Silver Bow Creek. For example, although channelization, land uses, temperature, nutrients, and dissolved oxygen can cause trout population reductions, these factors are inconsistent with observed data because:

- ▶ They do not explain observed fish kills and the results of *in situ* bioassays, both of which demonstrate ambient acute toxicity in the Clark Fork River/Silver Bow Creek caused by hazardous metals.
- ▶ They do not explain the results of food-chain growth reduction injuries caused by consumption of contaminated invertebrates collected from the Clark Fork River.
- ▶ They do not explain observed physiological impairment injuries in field-collected trout, which were similar to physiological impairment injuries observed in trout exposed to hazardous metals in controlled laboratory exposures.
- ▶ They are inconsistent with observed temporal patterns of injury. For example, conditions of aquatic biota in the Clark Fork River improved substantially following elimination of elevated metals discharges from the Weed Concentrator in the early 1970s and 1980s. Similarly, the frequency of observed fish kills in the Clark Fork River has decreased since removal of hazardous substance-contaminated tailings materials in the Mill-Willow Bypass. These events represent situations in which biological conditions have improved following removal of sources and reductions in metals inputs to the Clark Fork River; they were not associated with substantial changes in land uses, channelization, nutrients, temperature, or other ARCO hypothesized causes of trout population reductions.
- ▶ They are inconsistent with the observed differential degree of biological injuries in Silver Bow Creek and the Clark Fork River. Trout populations have effectively been eliminated in Silver Bow Creek (which contains the highest concentrations of metals in water and sediment), whereas trout are found in the Clark Fork River (which contains lower concentrations of metals in water and sediment than Silver Bow Creek). However, channelization and land uses do not differ markedly between Silver Bow Creek and the Clark Fork. Temperatures in Silver Bow Creek may be somewhat



**Table 6-2**  
**Causality Evaluation: Trout Population Reductions,**  
**Clark Fork River/Silver Bow Creek, MT**

	Plausible Cause of Observed Environmental Effect?					
	Channelization	Land Use	Nutrients	Temp.	D.O.	Hazardous Subs.
Can cause population reductions?	Yes	Yes	Yes	Yes	Yes	Yes
Consistent with observed temporal trends and conditions?	No	No	No	No	No	Yes
Explains differential level of injury to CFR and SBC?	No	No	No <sup>1</sup>	Yes <sup>2</sup>	No <sup>1</sup>	Yes
Evidence that lethality observed in <i>in situ</i> testing caused by...?	No	No	No	No	No	Yes
Evidence that fish kills caused by...?	No	No	No	No	No	Yes
Explains food-chain injuries from consumption of contaminated CFR invertebrates?	No	No	No	No	No	Yes
Consistent with health impairment injuries in field-collected fish?	No	No	No	No	No	Yes <sup>3</sup>
<sup>1</sup>	Are not plausible causes of trout population reductions because Butte Wastewater Treatment Plant effluent discharges which would be regulated absent contamination with hazardous substances (Kerr, Appendix A). Metals currently are limiting trout populations in Silver Bow Creek.					
<sup>2</sup>	Caused by absence of streamside vegetation, which has resulted from contamination of streamside soils with hazardous substances (see Lipton et al., 1995b).					
<sup>3</sup>	Consistent health impairment injuries were observed in field-collected trout and in trout exposed to hazardous metals in controlled laboratory testing (see Lipton et al., 1995a).					

elevated relative to the Clark Fork River; however, this is primarily a function of the elimination of shading vegetation because of contaminated, phytotoxic streamside tailings (see Lipton et al., 1995b). Nutrient and dissolved oxygen levels are not limiting in Silver Bow Creek because hazardous substances presently are limiting (Appendix A). Butte Wastewater Treatment Plant discharges would have been regulated by the State absent the substantial contamination of the creek with hazardous substances and will be controlled once the limiting factor (i.e., hazardous substances) is addressed (Kerr, Appendix A).

This analysis demonstrates that hazardous substances represent the only plausible, temporally and spatially coherent, and consistent explanation of observed trout population reductions; causal mechanisms proposed by ARCO do not.





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## CHAPTER 7

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**APPENDIX A**  
**RESPONSE TO ARCO'S REPORTS CONCERNING THE STATE OF**  
**MONTANA'S INJURY ASSESSMENT FOR THE UPPER CLARK FORK**  
**RIVER BASIN**



**RESPONSE TO ARCO'S REPORTS  
CONCERNING THE STATE OF MONTANA'S  
INJURY ASSESSMENT FOR THE UPPER CLARK FORK RIVER BASIN**

by

**Mark A. Kerr  
Natural Resource Damage Litigation Program  
Montana Department of Justice  
Old Livestock Building  
1310 East Lockey Avenue  
P.O. Box 210425  
Helena, Montana 59620-1425**

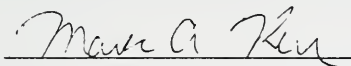


**RESPONSE TO ARCO'S REPORTS  
CONCERNING THE STATE OF MONTANA'S  
INJURY ASSESSMENT FOR THE UPPER CLARK FORK RIVER BASIN**

*Prepared by*

**Mark A. Kerr**

*Testifying Expert:*

A handwritten signature in cursive script, reading "Mark A. Kerr", is written over a horizontal line.

**Mark A. Kerr  
Natural Resource Damage Litigation Program  
Montana Department of Justice  
Old Livestock Building  
1310 East Lockey Avenue  
P.O. Box 210425  
Helena, Montana 59620-1425**

*Date:*

**October 13, 1995**



## INTRODUCTION

The purpose of this report is to respond to assertions made in part or in whole by various experts of ARCO concerning specific aspects of the State of Montana's Aquatics Resources Injury Assessment Report. Specifically, the themes or criticisms to be addressed are the following:

1). Silver Bow Creek would not presently support a trout fishery, in the absence of hazardous substances, due to adverse impacts from the Butte Wastewater Treatment Plant discharge;

2). adverse impacts from placer mining on Silver Bow Creek, on tributaries to Silver Bow Creek and the Clark Fork River, and on many other streams within the Clark Fork River basin, have resulted in detrimental (and potentially injurious) impacts to fisheries of Silver Bow Creek and the Clark Fork River; and,

3.) hardness concentrations used in fish toxicology studies conducted on behalf of the aquatics resources injury assessment were inappropriate because they were not representative of conditions in the Clark Fork River.

ARCO's experts who, alone or in concert with other experts, touch upon these various issues include Dr. Dudley W. Reiser; Dr. Andy Davis; Dr. Thomas C. Ginn; and Dr. Kenneth D. Jenkins.

## **IMPACTS OF THE BUTTE WASTEWATER TREATMENT PLANT DISCHARGE ON SILVER BOW CREEK**

A significant point of disagreement between the State and ARCO concerns the condition of Silver Bow Creek absent the release of hazardous substances. ARCO asserts that even if hazardous substances were not present in Silver Bow Creek, adverse impacts related to the Butte Wastewater Treatment Plant (WWTP) discharge would prevent the reestablishment of a trout fishery. In short, the "baseline" condition of Silver Bow Creek would be a trout population of zero. This would not be the case. If hazardous substances were not present in Silver Bow Creek, the Creek today would support a viable trout population.

ARCO, in its Report of Assessment, discusses the detrimental impact the discharge from the Butte WWTP has on surface water quality and salmonid populations in Silver Bow Creek. Ginn (1995), in his report on Surface Water Resources, presents an analysis of nutrient, ammonia and dissolved oxygen concentrations in Silver Bow Creek and exceedences of their respective criteria. Reiser asserts in his report (1995) (p. D4-48) that "Discharges from the Butte WWTP have historically affected both the water quality and the aquatic communities of this section of Silver Bow Creek...Exceedences in ammonia and nutrient releases and BOD loading have and continue to occur...These factors (nutrients, algae blooms, diurnal sags in dissolved oxygen levels, and ammonia) continue to preclude salmonid populations from reestablishing above the Warm Springs Ponds." The State quite simply disagrees with this assertion. Ammonia, dissolved oxygen and nutrients are potentially limiting factors in the future once metals' contamination is addressed. If this is the case, then the State and the U.S. Environmental Protection Agency (EPA) will require additional treatment of the Butte WWTP effluent in order to address the parameters that are limiting or potentially limiting to aquatic life. This would be consistent with actions taken at other facilities where adverse impacts to aquatic life related to the quality of a wastewater treatment plant discharge were documented or anticipated.

It is useful to briefly summarize the situation on Silver Bow Creek, both historically and as it is today, to understand the State's position. As early as 1907, the Montana Revised Codes, Section 1564, Chapter 177 prohibited the dumping of sewage, drainage, refuse, or polluting matter of any kind into any spring, pond, lake or stream used as a source of water or ice supply by the city, town, or public institution (HRC, 1993a). This regulation gave the State Board of Health the power to prevent pollution of waterways in the state. Even before this time, however, the city of Butte undertook various improvements to its sewerage collection and treatment system. By 1890 around 3.5 miles of sanitary sewers were completed. A number of documents point to continued growth of both storm and sanitary sewer lines in various parts of the city between 1890 to about the time of World War I (HRC, 1993a). By 1897, the sanitary sewage system consisted of 13 miles of lines. Various newspaper accounts describe further expansion of sewerage facilities in 1907, 1908 and 1909.

In 1911, the 1907 legislation was amended to require the State Board of Health to prove that the



sewage from a particular municipality was harming the downstream users before it could be prohibited. This action occurred in response to considerable opposition met by the State Board of Health while trying to enforce the law concerning the pollution of streams. Nonetheless, the Butte city council created the Silver Bow Creek Commission in 1923 to deal with "the work which may from time to time be required in order to keep [SBC] in a safe and sanitary condition" (HRC, 1993a).

There has been little documentation of what growth and changes occurred in the Butte sewerage system in the 1920s. But, even after the substantial extension of the storm and sanitary sewer systems, the untreated discharge went straight into Silver Bow Creek. In 1930, H.B. Foote, who was referred to as the State Board of Health director, noted the pollution caused by both domestic sewage and industrial wastes in Montana, using Silver Bow Creek as an example.

The development and maintenance of the Butte sewer system in the 1930s was affected by the New Deal programs. Under this program the city carried out numerous and extensive projects. Attempts to correct the on-going problems with the drainage (including noxious odors, deposition of sediments, and flooding) and to expand the sewer system in Butte continued during the 1940s. The idea of constructing some sort of interceptor for the sewage to rid Silver Bow Creek of the sanitary sewage discharge began to take form toward the end of the 1940s.

The question of solving the problem with Butte's sewage treatment was temporarily put to rest when the state issued the Montana Water Pollution Act of 1955 (HRC, 1993a). This law covered prevention, control and abatement of water pollution (Brinck et al., undated), and required that streams in the state be classified. The first streams were classified in 1958 beginning with the Yellowstone River, which was followed shortly thereafter by the Clark Fork and other major rivers. Most stream classification was completed in 1960 (Brinck et al., undated). The act and subsequent revision were increasingly criticized, however, because of the way in which Silver Bow Creek was allowed to continue to be used for industrial waste disposal (HRC, 1993b). The Act made any stream used for industrial waste for 30 years unable to be classified by the state for anything other than "industrial use only" (HRC, 1993b). In 1967 when this act was revised by the state legislature, the Silver Bow Creek classification remained the same.

Whether Silver Bow Creek between Butte and Warm Springs was a polluted stream or an open sewer continued to be debated during the 1950s. In a report prepared by C.W. Brinck for the Board of Health in March 1957 he noted that the pollution between Butte and Warm Springs was considered "gross." Through subsequent revisions of the State's water quality standards and reviews of stream classification, Silver Bow Creek moved to an "I" classification, which included streams that historically had severe water quality problems. Besides Silver Bow Creek, three streams three streams impacted by agricultural practices (Prickly Pear Creek, Hot Springs Creek and Muddy Creek) were also included in the "I" classification.

Despite the "unclassified" and "I" classification adopted for Silver Bow Creek since 1955, actions did occur over the years to improve the collection and treatment of sewage in the city of Butte. Not much documentation has been discovered regarding the Butte sewer disposal system between 1957 and 1965. When the Anaconda Company constructed its concentrator the neutralizing effects of the mine waters diminished, enabling fecal bacteria to survive in the stream and creating the need for a domestic sewage treatment facility. In August, 1965 the Silver Bow county commissioners adopted a resolution to create Metro Sanitary Storm and Sewer District No. 1 to construct interceptor sewers, repair existing sewers, and provide sewage treatment in the city of Butte and surrounding areas. The treatment plant was finished in 1969. In June 1970 a secondary treatment plant was built (although recovered sludge continued to be discharged into Silver Bow Creek until 1975). With passage of the Water Pollution Control Act Amendments in 1972, it became mandatory for all municipal wastewater discharges to receive a minimum of secondary treatment (MDHES, 1978). Butte's facilities, by this time, were already providing secondary treatment. Due to Silver Bow Creek's classification, however, additional treatment beyond conventional secondary treatment has to date been unnecessary. Clearly, as the quality of Silver Bow Creek improves due to the control of tailings dispersed across the Silver Bow Creek floodplain, the Butte WWTP will be required to upgrade the treatment of wastewater discharged to Silver Bow Creek. Language in the State's surface water quality standards (MDHES, 1988) makes this clear:

(Administrative Rules of Montana (ARM) 16.20.623: "I Classification (1): The goal of the state of Montana is to have these waters fully support the following uses: drinking, culinary, and food processing purposes after conventional treatment; bathing, swimming and recreation; growth and propagation of fishes and associated aquatic life, waterfowl, and furbearers; and agricultural and industrial water supply..."

ARM 16.20.623 (2)(h)(ii): "As the quality of these waters improves due to control of nonpoint sources, point-source dischargers will be required to improve the quality of their discharges following the MPDES rules (ARM Title 16, chapter 20, subchapter 9)."

It is useful to review actions at three other cities in Montana to demonstrate the State's and EPA's concern with water quality impacts associated with municipal wastewater discharges. These three case studies demonstrate that any adverse impacts in Silver Bow Creek due to the Butte WWTP discharge would have been addressed had it not been for the Creek's "I" classification. These studies involve two situations. In the first, it was determined during the planning or design of two WWTPs (Bozeman and Kalispell) that existing or potential adverse impacts to their receiving waters required additional enhanced wastewater treatment. In the second, it was determined that potential adverse impacts from ammonia at Helena needed to be addressed after the receiving water (Prickly Pear Creek) was reclassified for aquatic life use. These three case studies are summarized below.

Bozeman: In February, 1980 EPA issued a Finding of No Significant Impact (FONSI) for the Bozeman Wastewater Treatment Facility (EPA 1980). An Environmental Assessment (EA) attached to the FONSI described the project and analyzed impacts in detail. The city was being required to provide treatment greater than secondary because "the city and water quality experts explained the detailed wasteload allocation and fisheries studies that have been completed on the East Gallatin River...showed ammonia and chlorine levels too high to meet water quality standards." As stated in the EA:

"With the present discharge from the Bozeman WWTP, the quality of the East Gallatin is less than the B-D<sub>2</sub> [the same as the present B-2 classification] water use classification. Significant oxygen depletion is evidenced during warm temperature months. Nutrient concentrations below the discharge are significantly increased. Diurnal DO fluctuations indicate that significant photosynthetic activity is taking place downstream from the discharge. Unionized ammonia levels exceed the recommended level of 0.02 mg/l NH<sub>3</sub>-N during low flow stream conditions (unionized ammonia is toxic to aquatic life in sufficient concentrations). These high levels of unionized ammonia have been shown to persist for as far as 6 kilometers downstream from the point of discharge.

...The city of Bozeman recognized the need for ammonia reduction in the East Gallatin; however, they contested the idea of having to construct both the secondary and advanced secondary treatment facilities immediately. The city's intent was to construct the secondary treatment facility, then after a period of studying the secondary treatment plant and analyzing the then available technology, design and construct the advanced secondary treatment facility. This approach was unacceptable to the EPA and the Montana Department of Health and Environmental Sciences (DHES). Facility planning regulations require that the alternatives must be able to meet all permit limits, including advanced secondary treatment."

Kalispell: in the mid-1980s, during facilities planning for upgrading the Kalispell WWTP, studies conducted by the Water Quality Bureau identified low dissolved oxygen (DO) levels in Ashley Creek, the receiving water for the Kalispell WWTP and a tributary of the Flathead River. This problem was identified following the release of a FONSI in August, 1984. The following was stated in the Amendment to the Environmental Assessment for Kalispell, Montana (EPA, 1988):

"Water quality studies made on Ashley Creek, the receiving stream for Kalispell's effluent indicated that instream dissolved oxygen (DO) levels were adversely affected by the wastewater. The discharge permit limits for biochemical oxygen demand (BOD), total suspended solids (TSS) and residual DO were lowered to protect the receiving stream thereby satisfying state water quality standards. Reevaluation of treatment alternatives led to recommendation of a treatment system similar to the original proposal yet capable of meeting the more stringent limits."

Further stated in the "Purpose and Need for This Action" was the following:

"The FONSI prepared in August 1984 provided for construction of an advanced treatment facility to meet national secondary limits for BOD and TSS...Subsequent evaluation of the receiving stream indicated that additional controls on oxygen consuming pollutants would be necessary to maintain the water quality standard established for dissolved oxygen. New MPDES discharge permits required the City of Kalispell to reevaluate treatment alternatives."

Helena: the Helena WWTP discharges to Prickly Pear Creek. In the early 1990's, a study of Prickly Pear Creek concluded that ammonia in the Helena WWTP discharge was elevating instream concentrations of ammonia above criteria concentrations, and that a measurable impact to the benthic community existed.

The following language appears in the Statement of Basis (MDHES, 1992) for the City of Helena wastewater discharge permit:

"The Water Quality Standards classify the creek (i.e. the receiving water Prickly Pear Creek) as "I". The Standards state that the "goal" for this classification is to have these waters fully support the beneficial uses described in the B-1 classification...As the classification is likely to change, the toxicity requirements will be assigned based on a non-"I" classification...Based upon the "Prickly Pear Creek Ammonia Study Report", July 23, 1992 ammonia discharging from the Helena STP is exceeding Gold Book criteria. Therefore, this new permit will require ammonia monitoring of the effluent in the plant discharge and ditch, and in the creek. The purpose of this monitoring will be to further evaluate the ammonia impact on the creek and to provide the needed data when an ammonia effluent limitation is required in the future to meet the "goal" of the Standard's "I" classification.

At the present time, the city of Helena is beginning a facilities plan to upgrade wastewater treatment facilities (pers. comm., Tom Slovarp - Water Quality Bureau).

### Summary

The case studies described above illustrate that in situations where existing or potential adverse instream impacts related to WWTP discharges have been identified, the State and EPA have addressed or are addressing these impacts. In summary, it is inconceivable that absent the mining wastes and hazardous substances in and along Silver Bow Creek the State and EPA would have allowed discharges from the Butte WWTP to degrade the creek to the point where a trout fishery could not exist.



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## EFFECTS OF PLACER MINING ON AQUATIC RESOURCES

In the expert reports of Terry McNulty and Andy Davis, ARCO describes the extent to which placer mining has released sediments into the Clark Fork River and its tributaries, including Silver Bow Creek. The implication of the McNulty and Davis reports is that placer mining impacts for which ARCO claims it is not responsible are significant factors in the existing condition of aquatic resources in Silver Bow Creek and the Clark Fork River.

For example, Davis (pp. 1.7) notes spikes in metal concentrations in sediments just downstream and in the vicinity of the Clark Fork's confluence with the Little Blackfoot River, Willow Creek and Gold Creek, streams that have experienced historic placer and lode mining. Davis (Section B.2.f) also concludes that based on mass balance calculations of metals around the Gold Creek and Flint Creek tributaries, it is apparent that there is a contribution from historical lode and placer mining on these creeks to the Clark Fork mainstem. Davis (pp 1.10 to 1.11) also states that extensive hardrock and placer mining on many tributaries to the Clark Fork have resulted in the transport of metals downstream to the Clark Fork. McNulty (p. 4) concluded that placer mining during the period of 1860 - 1947 caused the movement and redeposition of 55,000,000 tons of sediments in the Clark Fork River and its tributaries.

While it is true that placer mining released sediments to surface waters downstream of placer mining operations, and that physical impacts to stream channels and fish habitat are evident even today from historic placer mining activity on many streams in the Clark Fork Basin, it cannot be concluded that historic placer mining would have resulted in the elimination or even a substantial reduction in fish populations on Silver Bow Creek or the Clark Fork River today. This is true for the following reasons:

- 1.) Silver Bow Creek today exhibits comparatively little physical evidence of historic placer mining compared to many other streams in Montana which have been placer mined, indicating that permanent destruction of trout habitat due to placer mining is minimal;
- 2.) many streams that contain viable trout populations have been placer mined and exhibit more extensive physical impacts to the stream channel than Silver Bow Creek;
- 3.) reference streams for fish injury quantification (Flint Creek, Big Hole River, Rock Creek and Ruby River) also have had extensive historic placer mining activity on their tributaries. If one accepts the "river continuum" concept endorsed by ARCO (Reiser, p. C-28), then the State's reference streams, as well as many other productive trout streams in the State that have experienced placer mining in their tributaries, are similarly impacted by historic placer mining. Consequently, differences in trout populations between Silver Bow Creek, the Clark Fork River and the State's reference streams cannot be attributable to effects of historic placer mining because the State's reference streams have also been affected by placer mining.

### Comparison of Silver Bow Creek to other creeks

Much of the remaining physical evidence of historic placer mining on Silver Bow Creek occurs along the reach from approximately Rocker to Ramsay (GCM, 1992). The streamchannel in this reach is relatively well-defined, and little evidence of physical disturbance (placer tailings, mounded alluvial gravels) exists. GCM (1992) notes "Many of the early placer features have been degraded by subsequent placer mining, the erosive action and deposition of materials by Silver Bow Creek, and railroad construction." By comparison, many other streams in State still exhibit severe physical impacts to their streamchannels and floodplains. It is evident from the photographs that these streams have more physical disturbance associated with historic placer mining activities than has Silver Bow Creek. While the amount of physical disturbance associated with historic placer mining can range from minimal to severe, Silver Bow Creek exhibits relatively few impacts compared to other placer-mined streams in Montana.

### Trout populations in placer-mined streams

Although placer mining can result in substantial physical impacts to a streamchannel and severely degrade habitat components necessary for a trout fishery, placer mining alone does not necessarily result in the elimination of a trout fishery. Table 1 summarizes trout population information for streams that have been altered by placer or dredge mining. The physical effects from placer mining on these streams (other than Silver Bow Creek) are quite significant and readily apparent to this day.

Table 1. Trout Populations in Streams Affected by Historic Placer Mining

Stream	Tributary to:	Trout Species Present	Number trout per mile <sup>1</sup>
Prickly Pear Creek	Missouri River	rainbow, brown, brook	410 <sup>2</sup>
Silver Creek	Missouri River	cutthroat	present <sup>3</sup>
Lowland Creek	Boulder River	brook, rainbow	1,779 <sup>4</sup>
Grasshopper Creek	Beaverhead River	brook, brown, rainbow	58 <sup>5</sup>
German Gulch	Silver Bow Creek	brook, cutthroat	1,067 <sup>6</sup>
Clancy Creek	Prickly Pear Creek	brook	174
Silver Bow Creek	Clark Fork River	none	0

1. Fish population estimates provided where available. Otherwise, qualitative information provided.

2. La Point et al., 1982.

3. Phillips, G. Personal Communication.

4. MDFWP, 1995.

5. MDFWP, 1981.

6. Reiser, 1995.

7. MDFWP, 1984.







Figure 1. Placer mining impacts in German Gulch.





Figure 2. Placer mining impacts on Prickly Pear Creek.





Figure 3. Placer mining impacts on Lowland Creek.







Figure 4. Placer mining impacts on Clancy Creek.





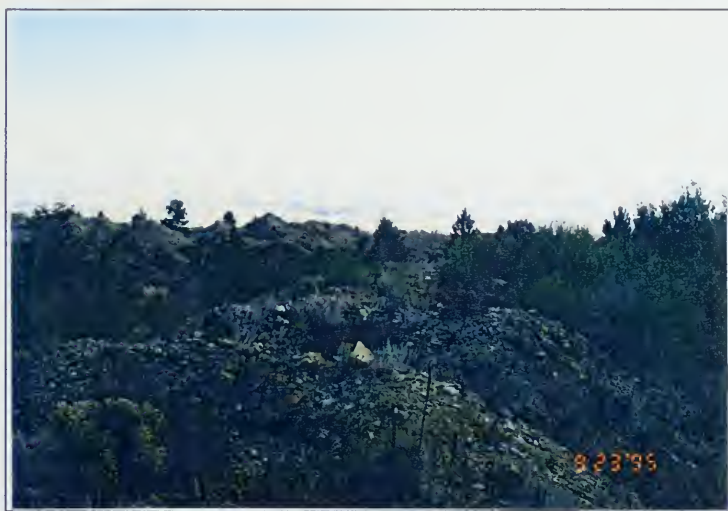


Figure 5. Placer mining impacts on Silver Creek.





Figure 6. Placer mining impacts on Silver Bow Creek.



### Placer mining within reference stream drainages

As noted above, placer mining has occurred on many streams tributary to the State's fish population reference streams. Consequently, the State's baseline fish estimates have accounted for and integrated the effects of placer mining. The following streams were identified as having had placer mining activity (Lyden, 1948). It should be noted that the streams identified below supported a range of placer mining activities from small scale prospecting and sporadic mining to intensive dredging operations that lasted for many years. Consequently, the degree of physical alteration of these streams ranges from minimal to severe.

#### Rock Creek Drainage:

Basin Creek  
Quartz Creek  
Big Springs Creek  
Welcome Creek  
Brewster Creek  
Upper Willow Creek

#### Flint Creek Drainage:

Flint Creek mainstem  
Henderson Creek  
Princeton Gulch  
Little Gold Creek

#### Big Hole River Drainage:

Rochester Creek	McCarthy Creek
Moose Creek	Soap Creek
Camp Creek	Divide Creek
French Creek	California Creek
Oregon Creek	Mulchey Creek
Ruby Creek	Trail Creek
May Creek	Placer Creek

#### Ruby River Drainage:

Basin Creek	Poison Creek
East Fork Ruby River	Burnt Creek
Cottonwood Creek	Lazyman Creek
Warm Springs Creek	South Fork Warm Springs Creek
Middle Fork Warm Springs Creek	French Creek
Greenhorn Creek	Idaho Creek
Barton Creek	Alder Gulch
Harris Creek	California Creek
Ramshorn Creek	Wisconsin Creek
Bivens Creek	

Beaverhead River Drainage:

Rattlesnake Creek  
Grasshopper Creek  
Jeff Davis Gulch  
Maiden Creek

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## HARDNESS CONCENTRATIONS IN THE CLARK FORK RIVER

ARCO has been critical of the test conditions under which several of the State's fish toxicology studies have been conducted, questioning whether these conditions are representative of conditions in the Clark Fork River. The hardness component of the studies' test conditions has been particularly criticized. ARCO contends that the 100 mg/l hardness concentration employed in the various studies is substantially less than ambient hardness concentrations in the Clark Fork River. In his Expert Report, Kenneth Jenkins (p. 6) states "Water hardness levels employed in the laboratory studies were substantially lower than those reported in the Clark Fork River..". Jenkins further comments (p. 22) that "...the hardness levels in the laboratory studies were generally in the 95 to 125 mg/l range...with the exception of one test in the pulse exposure studies in Appendix B, the hardness level values for the laboratory toxicity tests were consistently less than 50% of those hardness level values observed in the Clark Fork River." The statement implies that a hardness concentration of at least 190 to 250 mg/l would be more appropriate for the toxicology studies than the 100 mg/l nominal concentration employed by the State.

As a preliminary matter, it must be emphasized that test conditions employed in the fish toxicology studies were intended to represent spring conditions in the Clark Fork River. While Jenkins' implied hardness concentrations of 190 to 250 mg/l are generally quite typical of conditions in the Clark Fork River for much of the year, they are definitely not typical of conditions during the spring that the fish toxicology studies were intended to simulate. In spring, hardness concentrations are generally at their lowest levels of the year because of dilution related to snowmelt. Metals concentrations also tend to be at the highest concentrations of the year due to erosion of contaminated floodplain and streambank sediments, and resuspension of contaminated streambed sediments. The combination of low hardness levels and high metals concentrations are potentially the most toxic conditions during the year to which trout are exposed and may represent a bottleneck in the viability of trout populations. Although there is year-to-year variability in the timing, duration and intensity of this phenomenon, it occurs every spring throughout the Clark Fork River and its tributaries. In general, hardness concentrations which commonly exceed 250 mg/l in much of the Clark Fork River during a substantial portion of the year frequently fall into the 95 to 125 mg/l range during spring runoff.

To demonstrate the appropriateness of using a hardness concentration of 95 to 125 mg/l in the State's fish toxicology studies, hardness data from three sites on the Clark Fork River were evaluated for the years 1989 to 1994. These sites (Deer Lodge, Gold Creek and Turah) encompass virtually the entire length of the river. The analyzed data were collected in the months (April, May and June) that coincide with the major portion of spring runoff. The minimum hardness concentrations for these three months for the years 1989 to 1994 are presented in Table 1.

Minimum hardness concentrations are mostly within the range of 95 to 125 mg/l. The data demonstrate the spatial variability of hardness within the Clark Fork River. Hardness is highest in the upper portion of the river downstream of Warm Springs Ponds and is lowest in the lower portion of the river downstream of its confluence with Rock Creek. The data indicate that while hardness concentrations in the upper river (Deer Lodge) are somewhat higher than the upper range of test

conditions (by about 30 mg/l on average), concentrations in the lower river (Turah) are about 10 mg/l less than the lower end of the range of test conditions, on average. For the entire Clark Fork River minimum hardness concentrations for every year except 1992 are within the range of test conditions.

In summary, the range of hardness conditions utilized in the fish toxicology studies are representative of conditions that occur in the spring in the Clark Fork River and are appropriate as test conditions. The range of hardness concentrations implied by Jenkins would be entirely inappropriate as test conditions attempting to simulate spring conditions in the Clark Fork River.

Table 1.  
Minimum Hardness Concentrations (mg/l)

Year	Deer Lodge	Gold Creek	Turah	Clark Fork River <sup>1</sup>
1989	150	141	76	122
1990	175	121	80	125
1991	109	113	74	99
1992	203	ND <sup>2</sup>	105	154
1993	150	123	73	115
1994	152	122	89	121
1989-1994	109	113	73	99
<sup>1</sup> . Values in this column are an average of values for the Deer Lodge, Gold Creek and Turah sites. <sup>2</sup> . ND (no data collected for this site in 1992).				







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**APPENDIX B**  
**EVALUATION OF THE CHRONIC TOXICITY OF CLARK FORK RIVER**  
**INVERTEBRATES TO RAINBOW TROUT (*ONCHORHYNCHUS MYKISS*)**



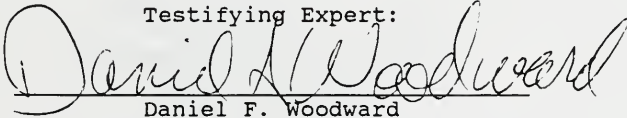
Report of Review of ARCO  
Document 0480-272

EVALUATION OF THE CHRONIC TOXICITY OF CLARK FORK RIVER  
INVERTEBRATES TO RAINBOW TROUT (ONCORHYNCHUS MYKISS) WHEN  
ADMINISTERED BY THE DIET

Prepared by:

Daniel F. Woodward

Testifying Expert:

A handwritten signature in black ink, reading "Daniel F. Woodward". The signature is written in a cursive style with a large, looping initial "D".

Daniel F. Woodward  
National Biological Service  
Midwest Science Center  
Jackson, Wyoming 83001

Date:

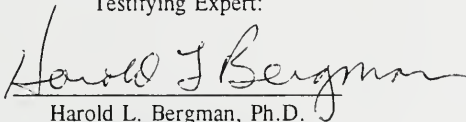
October 12, 1995



Report of Review of ARCO  
Document 0480-272

EVALUATION OF THE CHRONIC TOXICITY OF CLARK FORK RIVER  
INVERTEBRATES TO RAINBOW TROUT (ONCORHYNCHUS MYKISS) WHEN  
ADMINISTERED BY THE DIET

Testifying Expert:

A handwritten signature in black ink, reading "Harold L. Bergman". The signature is fluid and cursive, with the first name "Harold" and last name "Bergman" clearly legible. The signature is written over a horizontal line.

Harold L. Bergman, Ph.D.  
Department of Zoology and Physiology  
University of Wyoming  
Laramie, Wyoming 82071

Date:

October 12, 1995





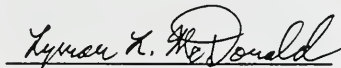
Report of Review of ARCO  
Document 0480-272

EVALUATION OF THE CHRONIC TOXICITY OF CLARK FORK RIVER  
INVERTEBRATES TO RAINBOW TROUT (ONCORHYNCHUS MYKISS) WHEN  
ADMINISTERED BY THE DIET

Prepared by:

Daniel F. Woodward

Testifying Expert:

A handwritten signature in cursive script, reading "Lyman L. McDonald". The signature is written in dark ink and is positioned above a horizontal line.

Lyman L. McDonald, Ph.D.  
WEST, Inc.  
2003 Central Avenue  
Cheyenne, Wyoming 82001



Report of Review

Atlantic Richfield Company  
Document 0480-272

EVALUATION OF THE CHRONIC TOXICITY OF CLARK FORK RIVER  
INVERTEBRATES TO RAINBOW TROUT (ONCORHYNCHUS MYKISS) WHEN  
ADMINISTERED BY THE DIET

Studies Conducted By  
ENSR Consulting and Engineering

Authors

Daniel F. Woodward  
National Biological Service  
Midwest Science Center  
Jackson, Wyoming 83001

Harold L. Bergman  
Department of Zoology and Physiology  
University of Wyoming  
Laramie, Wyoming 82071-3166

Lyman L. McDonald  
Western EcoSystems Technology, Inc.  
Cheyenne, Wyoming 82001

Contributing Author

Aida M. Farag  
National Biological Service  
Midwest Science Center  
Jackson, Wyoming 83001

## INTRODUCTION

The purpose of our review of the ENSR Consulting and Engineering research report (ENSR 1995) was to interpret the findings on food-chain toxicity, so that we can further our understanding of the role of metals contamination of aquatic invertebrate food-chains and its effect on reducing health of early life stage trout in the Clark Fork River. We believe the ENSR findings are important, if properly interpreted and analyzed, and provide significant additional information. Following is a review of conditions present in the Clark Fork River and previous research which has led up to the current experiments performed by ENSR.

The upper Clark Fork River has been well characterized for its extensive trace metal contamination in both abiotic and biotic components (Luoma et al. 1989; Moore and Luoma 1990; Axtmann and Luoma 1991; Cain et al. 1992). Benthic organisms in the upper Clark Fork River have recently been implicated as a dietary source of metals that may be a chronic problem for young-of-the-year rainbow trout (Oncorhynchus mykiss) and brown trout (Salmo trutta) (Woodward et al. 1994, 1995). Both species were exposed for 88-91 d to simulated Clark Fork River water and a diet of benthic invertebrates collected from the river. These exposures resulted in reduced growth and elevated levels of metals in the whole body of both species and in the livers of rainbow trout. Brown trout fed on the metals-contaminated diets exhibited constipation, gut impaction, increased cell membrane

damage (lipid peroxidation), decreased digestive enzyme production (zymogen), and a sloughing of intestinal mucosal epithelial cells. Rainbow trout fed the contaminated diets exhibited constipation and reduced feeding activity. Similar physiological impairments have been noted in fish collected from the field (Farag et al. 1995), and it is generally accepted that the reduced standing crop of trout in the Clark Fork River (Johnson and Schmidt 1988; Chapman 1993) resulted partly from chronic effects of metals contamination in benthic invertebrates that are important as food for young-of-the-year fish (Woodward et al. 1994, 1995).

The food-chain effect of metals has been described by others as a relationship in which biomagnification is not observed and bioconcentration factors are small, but the amount of metal transferred by food can be high enough to attain biologically harmful concentrations in fish (Dallinger et al. 1987). Age-0 brown trout and rainbow trout are more susceptible than older fish because their diets consist totally of drifting benthic invertebrates and zooplankton (Carlander 1969; Hubert and Rhodes 1992). Newly hatched rainbow and brown trout initially feed on zooplankton but soon shift to small dipterans and ephemeropterans.

The significance of food-chain uptake of metals by fish has been observed in other aquatic systems where there was substantial metals contamination of sediments, macrophytes, and benthic invertebrates (Patrick and Loutit 1978; Dallinger and

Kautzky 1985; Dallinger et al. 1987; Harrison and Klaverkamp 1989). Investigators have stressed the importance of using environmentally contaminated natural foods (Harrison and Curtis 1992) and the specific ecological situation of a given environment (Dallinger et al. 1987) in assessing effects of contaminants on fish populations.

Earlier experiments performed by ENSR (Mount et al. 1994) controlled noncontaminant dietary factors in the laboratory, but conditions were unnatural and did not simulate the Clark Fork River. The Mount study used brine shrimp (Artemia), an invertebrate source not found in the Clark Fork River. Artemia were exposed to an aqueous metals mixture for less than 24 h after hatching, and then fed to rainbow trout. Metals concentrations in Artemia were elevated to concentrations comparable to our field collected Warm Springs (WS) diet (Woodward et al. 1995), but rainbow trout fed on the exposed Artemia did not show reduced survival or growth. However, the duration of Artemia exposure was so short that it is doubtful that normal metabolic assimilation of the metals occurred in the newly hatched brine shrimp. As a result, the metals attached to brine shrimp were likely in the free ionic form. The impacts of metabolized metals far outweigh that of simple ions (Hodson 1988) in the food-chain, and it is known that As, Cd and Cu are metabolized by aquatic biota (Craig et al. 1986). The absence of effects in the Mount et al. (1994) study was similar to those experiments where ionic metals were applied surficially to dry

diets (Wekell et al. 1983; Lanno et al. 1985; Crespo et al. 1986), but did not mimic conditions in the natural systems.

The current ENSR study (1995) was a dietary exposure study which controlled some of the variables criticized in the Mount and Woodward studies (Mount et al. 1994; Woodward et al. 1994, 1995). The ENSR (1995) study determined the effects of metals contamination in a live-natural freshwater diet by comparing survival and growth among groups of rainbow trout fry fed live, laboratory-reared Daphnia pulex and groups of rainbow trout fry fed live, planktonic invertebrates (primarily Daphnids and copepods) collected from the discharge at Warm Springs Ponds (WSP). Therefore, the WSP diet consisted of environmentally contaminated natural foods, of resident species in the Clark Fork Basin, and of a species composition which was standardized as much as possible between control and test diets. Not using natural environmental contamination processes or species present in the Clark Fork River were issues raised about the Mount (1994) study. Not having a standardized species composition in the test and reference diets was an issue which ENSR raised concerning the Woodward et al. (1994, 1995) studies.

## METHODS

The methods are reported in ENSR (1995) and will be briefly summarized.

Experimental design. Rainbow trout were exposed to six different treatment combinations of metals in water and diet.

There were three water exposures: 0X, 1X, and 2X; which corresponded to dissolved levels used in the Woodward et al. (1994, 1995) and Mount et al. (1994) experiments. The 1X water contained the ambient water criteria concentration (chronic) of Cd, Cu, and Pb, and was about 50% below criterion for Zn (USEPA 1987). These concentrations are also in the 75th percentile for dissolved measurements reported for the Clark Fork River by USGS between 1985 and 1990 (Lambing 1991). The nominal 1X metals concentrations (ug/L) were as follows: Cd, 1.1; Cu, 12; Pb, 3.2; and Zn, 50.

Metals measured in water were within acceptable ranges for the nominal concentration of each metal. However, Cu was elevated in the control (0X) water. Mean Cu concentration was  $8.4 \pm 0.8$  ug/L and should typically be less than 1 ug/L in control water.

There were two dietary invertebrate exposures: a test diet of resident invertebrates collected from the WSP discharge and a control diet (CON) of Daphnia pulex cultured in ENSR laboratory water. There was also an additional treatment consisting of fish fed a reference commercial trout diet (REF) in 0X water. Each of the seven treatments had four replications (see text table).



## Experimental Design

Diet	Water		
	0X	1X	2X
CON	X	X	X
WSP	X	X	X
REF	X		

Test Organism. The ENSR laboratory received rainbow trout for testing on May 18, 1994. This was 40 days after hatching, indicating the fry would have been on feed for approximately 20 days (water temperature, 10°C) prior to receipt by ENSR. Fry were held by ENSR for another 18 days until testing was initiated on June 5, 1994 (58 days post-hatch). From May 24 until June 5, all fry were fed the CON diet. Exposure was terminated after 46 days on July 21, 1994 when fish were 104 days post-hatch.

Diets and Feeding. The REF diet (Nelson's Sterling Silver Cup Fish Food) was formulated by Murray Elevators (Murray, Utah) to meet the specific nutritional requirements of trout diets (15% fat, 50% protein). The REF diet was fed by scoop prior to day 27, after which time aliquots of the REF diet were weighed. There were no partial feedings of the REF diet. The CON diet consisted of Daphnia pulex, which were cultured in mass under standard procedures in the ENSR laboratory. Invertebrates collected with plankton nets from the discharge of WSP were

predominantly Daphnia pulex and were used as the test diet. However, after the first three weeks, the dominance of D. pulex was inconsistent and species composition included copepods and cladocera. Fish were fed once per day, but on 5 days, partial feeding was necessitated by insufficient amounts of CON or WSP diets. However, on all but one occasion, CON and WSP fish were given equivalent amounts of feed through day 45. ENSR terminated the test on day 46 due to their claim of inconsistent species composition and insufficient feed supply.

See ENSR (1995) report for details on nutritional analysis, biological observations, wet chemistry, analytical chemistry for metals in diet, water, and fish, and statistical procedures.

## RESULTS AND DISCUSSION

Diet quality and metals concentration. In terms of fat, protein, ash, and energy available, the CON and WSP diets are similar (see text table).

Nutritional Content of Control (CON), Warm Springs Ponds (WSP), and Reference (REF) Diets (from ENSR 1995)

DIET	% FAT	% PROTEIN	% ASH	Energy <sup>1</sup>
CON	14.4	23.5	1.9	207
WSP <sup>2</sup>	16.2	25.3	2.1	228
REF	14	52	12	315

<sup>1</sup> kcal of energy available from fat and protein measured

<sup>2</sup> composite sample reported by ENSR (1995)

The similar nutritional contents of the CON and WSP diets could be expected because the protein source is daphnids, copepods, and

cladocera in both diets. The CON and WSP diets were collected under natural conditions, and the protein and fat contents of CON and WSP diets were not enhanced to maximize fish growth. The quantity of protein in the REF diet is twice that of the CON and WSP diets, and the protein source is derived from fish meal rather than invertebrates, which provides the best protein source for maximum growth and food conversion in fish. The REF diet is also formulated with the essential vitamins and minerals. Therefore, the REF diet is formulated commercially to maximize fish growth and the growth of fish on that diet should be evaluated as a "best case" diet condition. There are gross differences in the commercial Silver Cup diet (REF) and the CON and WSP diets in terms of quantity and quality of protein, energy availability, and presence of vitamins and minerals. Growth data from fish fed the REF diet cannot be compared to data from fish fed the CON and WSP diets. If we are trying to eliminate all variables except metals, the CON and WSP diets should only be compared with each other in terms of growth effects due to metals.

Except for Cd, mean concentrations of metals measured in invertebrates collected from WSP were 2 to 6.7 times lower than the concentrations of metals measured in invertebrates collected from the Clark Fork River (from Woodward et al. 1994, see text table).

Mean Concentrations of Metals (mg/kg, dry weight;  $\pm$  standard deviation) in various Invertebrate Diets; Control (CON) and Warm Springs Ponds (WSP) diets from ENSR (1995) Study, and in the Reference Snake River Invertebrate (SRI) and Clark Fork Invertebrate (CFI) diets from Woodward et al. (1994).

PARAMETER	CON	WSP	SRI	CFI
As	0.8 $\pm$ 0.9	20 $\pm$ 8.0	3.5 $\pm$ 1.0	43 $\pm$ 14
Cu	17 $\pm$ 11	61 $\pm$ 24	14 $\pm$ 3.1	381 $\pm$ 149
Cd	8.7 $\pm$ 2.3	4.0 $\pm$ 1.9	0.36 $\pm$ 0.36	3.1 $\pm$ 1.31
Pb	2.8 $\pm$ 2.2	4.9 $\pm$ 1.9	<2	33 $\pm$ 15

Cadmium concentrations were similar between WSP and CFI, but Cd concentrations were higher in the ENSR laboratory CON diet of Daphnia pulex than the reference SRI diet. The ENSR CON diet was actually 2-fold higher in Cd than the WSP diet. The source of elevated Cd in the ENSR laboratory diet is unclear and should be investigated.

Statistical Analysis of Growth Data. Because the REF diet was nutritionally different from the CON and WSP diets in the ENSR study, growth data of fish fed the REF diet should be excluded from statistical analyses; and inferences from these analyses should be limited to differences between the CON and WSP diets. These two diets are similar nutritionally and in species composition, but the concentrations of metals in the two diets are different and this difference is the issue under investigation. Also, inclusion of data from the REF diet in their statistical analysis promoted ENSR to use statistical procedures with reduced power compared to the standard procedures normally recommended for analysis of data from these types of studies. Thus, we conducted independent analysis of the growth

results reported in the ENSR study.

Inclusion of data from the REF diet in the analysis by ENSR (1995) causes a significant problem in the statistical analysis. The variables "weight" gain and "length" increase both have larger values for the REF diet than for the diets of primary interest. The problem in statistical analysis arises because of a corresponding increase in the variance of observations in the REF diet or with over-concern about "non-normality of the data". These combined data fail the test for equal variances or the test for normality of the data; therefore, ENSR (1995) used a statistical procedure (Kruskal-Wallis Analysis of Variance by Ranks followed by Dunn's Multiple Comparison-Kruskal-Wallis test) with reduced power to detect statistically significant differences. In this case, selection of a test with low power was to their clients advantage because significant differences would not be observed and biological effects would be minimized.

This problem can easily be addressed to allow one to use the more common and more powerful statistical tests. First, it is well known in the statistics literature that analysis of variance is reliable when randomization is used, regardless of the distributions involved (Kempthorne 1966). Concern with normality of the data is not a primary issue, because inferences are to means and means will approximately follow the normal distribution (a consequence of the central limit theorem (Sokal and Rohlf 1981)). Second, if data from the REF diet are dropped from the analysis, the remaining diets do not exhibit significant

departure from homogeneity of variances. Third, data (including the REF data) can be transformed to logarithms to eliminate significant unequal variances.

We analyzed the data with these three appropriate procedures and all three lead to substantially the same statistical conclusions. These conclusions are quite different than those reached by the less powerful technique selected by ENSR (1995). A brief review of our three analyses follows.

Two appropriate statistical analyses were conducted with the REF data eliminated. The first was based on the summary statistics in the ENSR (1995) report (Appendix A). We used appropriate two-sample t-tests with a Bonferoni correction for the 15 possible comparisons of the CON and WSP diets and the 0X, 1X, and 2X water treatments. The weight and length tables on the following pages summarize these results and a full account is in Appendix A. Significance of each comparison was indicated at a conservative level,  $\alpha=0.05/15=0.00333$ .

The second analysis was conducted on raw data provided to the State on September 20, 1995. With the REF treatment excluded, the design was reduced to a two-factor analysis of variance (Appendix B). Levene's test was used to test for homogeneity of within treatment variances (6 variances, combinations of 2 diets and 3 metal concentrations in water). For all three variables, the homogeneity of variances assumption was not rejected ( $\alpha=0.01$ ). Standard two-factor (diet and water) analysis of variance was conducted because the necessary



assumptions are satisfied. Tukey's method for multiple comparisons was used to test for significant differences between levels of the main effects, since the interactions for all three variables were not significant. Use of this method leads to substantially the same conclusions as reported when using the summary statistics and reported in the body of the paper (Appendix B for results).

A third analysis was conducted on raw data including the REF diet data. In this third analysis, we followed recommendations in the guidance document for Toxstat Version 3.4 software (West, Inc. And Gulley 1994) to transform the data if the data indicate significant departure from normality. All data values were transformed to logarithms to stabilize the variances and improve symmetry of the data. The homogeneity of variances assumption could not be rejected ( $\alpha=0.01$ ) for these transformed data and the follow-up analysis of variance leads to substantially the same conclusions as the two analyses reported in this report. Results of this third analysis, which supports our conclusions, are redundant and are not given herein.

Growth. Fish in the REF/OX exposure grew to 1.6 g during the 46 day exposure, and this growth rate could be considered optimum for rainbow trout under controlled conditions where the recommended amount of fat and protein is offered and feeding rate is not limited due to supply shortages. However, growth of fish fed the two invertebrate diets (CON and WSP) was less than half that of fish fed the commercial diet.

Growth of fish fed the WSP diet was significantly less over 46 days when compared to growth of fish fed the CON diet. Weight of fish fed the WSP diets, but exposed to different waters, were 76% to 69% of the size of those fish fed the respective CON diets (see text tables). Effects on fish length were similar to those on weight, but were less sensitive.

Weight (g) Differences after 46 days for fish on the Control (CON) and Warm Springs Ponds (WSP) Diets

CON DIET WT	WSP DIET WT	DIFF FROM CON	% OF CON
CON/0X 0.848	WSP/0X 0.625	-0.223	73.7% <sup>1</sup>
CON/1X 0.866	WSP/1X 0.643	-0.223	75.7% <sup>2</sup>
CON/2X 0.819	WSP/2X 0.564	-0.255	68.9% <sup>3</sup>

<sup>1</sup> Significant at P=0.0001

<sup>2</sup> Significant at p=0.0001

<sup>3</sup> Significant at p=0.0022

Length (mm) Differences after 46 days for fish on the Control (CON) and Warm Springs Ponds (WSP) Diets

CON DIET LT	WSP DIET LT	DIFF FROM CON	% OF CON
CON/0X 44±0.8	WSP/0X 40±0.7	-4.0	91 <sup>1</sup>
CON/1X 44±0.2	WSP/1X 41±0.2	-3.2	93% <sup>2</sup>
CON/2X 44±1.0	WSP/2X 39±1.1	-4.7	91% <sup>3</sup>

<sup>1</sup> Significant at P=0.0003

<sup>2</sup> Significant at p=0.0001

<sup>3</sup> Significant at p=0.0007

The growth effects observed were due to diet exposure (CON/0X versus WSP/0X, CON/1X versus WSP/1X, CON/2X versus WSP/2X) and not water exposure (CON/0X versus CON/1X and CON/2X; and WSP/0X versus WSP/1X and WSP/2X). There was a trend towards reduced growth in the 2X water (CON/0X versus CON/2X; and WSP/0X versus WSP/2X), but these differences were not significant.

The ENSR (1995) results are very similar to the Woodward et



al. (1994) study where exposure to metals through the CFI diet significantly reduced growth regardless of water exposure. After 21 d of feeding in the Woodward et al. (1994) study, rainbow trout fed the CFI diet were 15% smaller by weight than fish fed the SRI diet, and after 70 d of feeding, they weighed 37% less than the controls. Rainbow trout on the WSP diet in the ENSR study weighed about 27% less than the controls after 46 d of feeding. Furthermore in Woodward et al. (1995), brown trout weight reduction was 40% of the controls after 62 d on the WS diet; and rainbow trout weight was 20% of the control after 70 d of dietary (WS) exposure.

It is unfortunate that the ENSR study (1995) was terminated early after only 46 days of exposure. If the experiment could have continued through the planned 60 days, the magnitude of growth differences between the WSP and CON diet groups would have been greater, and growth curves could have been plotted. Also, the growth differences observed at day 46 would have been greater if the 46 days of food and water exposure would have occurred immediately after hatching instead of 58-d post hatch. Swim-up is the most sensitive life stage and is when exposure naturally occurs in the Clark Fork River. If the earliest life stage would have been exposed, less food would have been required, and the test could have continued for a longer period of time.

Even under the existing experimental design a few adjustments would have allowed continuation of the experiment. When lack of an adequate supply of laboratory reared Daphnia was

determined, the decision should have been made to eliminate either the 1X or 2X water exposure. This would have reduced the demand for Daphnia by one-third, allowing the study to continue. Changes in species composition of WSP invertebrates (Daphnia to copepods) was anticipated, and ENSR could have made the same species composition adjustment to the control diet in the laboratory.

While there were no objective measurements of feeding behavior, ENSR (1995) reported that rainbow trout preferred the Daphnia over the copepods because it took too much energy for the fish to pursue and capture the smaller copepods. While trout on both the WSP and CON diets received the same amount of diet, ENSR stated that fish on the WSP diet may have consumed less food because of the difficulty in capturing smaller prey.

There is another data-based explanation for the findings ENSR may have observed. Studies using objective feeding behavior measurements have shown that rainbow trout on metals contaminated diets from the Clark Fork River (WS and GC) had depressed feeding rate when compared to rainbow trout feeding on reference invertebrate diets (TB) (Woodward et al. 1995). In this study, both the test and control source of invertebrates were prepared into a dry pellet type diet. Therefore, when size of diet and ability to capture was uniform between control and test diets, feeding behavior was strongly affected by dietary metal concentration. The feeding activity of rainbow trout fed metals contaminated diets was less than 50% of fish fed the control

diet.

Metals in Fish Tissue. In the ENSR (1995) study arsenic, copper, and lead are elevated in tissue of fish from the WSP/2X treatment when compared to the controls (CON/0X) (see text table).

Mean Concentration (mg/kg, dry weight) of Metals in Fish tissue from various exposure sources: diet sources = Control (CON), Warm Springs Ponds (WSP), Snake River Invertebrates (SRI), Clark Fork Invertebrates (CFI), Warm Springs (W); water exposure = 0X, 1X, and 2X; 85th percentile for 112 stations sampled across the country in the National Contaminant Biomonitoring Program.

Source	As	Cd	Cu	Pb
CON/0X <sup>1</sup>	-	1.8	6.8	0.44
WSP/2X <sup>1</sup>	1.6	1.0	17	4.6
SRI/0X <sup>2</sup>	0.3	0.1	0.6	0.1
CFI/2X <sup>2</sup>	2.5	0.65	11	1.6
W/1X <sup>3</sup>	0.8	0.14	8.1	0.75
85th% NCBP <sup>4</sup>	1.1	0.2	4.0	0.88

<sup>1</sup> Data from ENSR 1995

<sup>2</sup> Data from Woodward et al. 1994

<sup>3</sup> Data from Woodward et al. 1995

<sup>4</sup> Data from Schmidt and Brumbaugh 1990; dry wt conversion, 4X

However, the tissue concentrations of Cd, Cu, and Pb in the controls (CON/0X) appear to be quite high when compared to the SRI/0X control and the 85th percentile values for the nation (Schmidt and Brumbaugh 1990). Both Cd and Cu in ENSR control fish (CON/0X) are higher than the 85th percentile values for 112 sampling stations across the country. The tissue Cd concentration of 1.8 mg/kg reported for CON/0X is 2-times higher than the maximum value of 0.9 mg/kg measured in fish throughout the national monitoring program. Cadmium concentrations were

also high in the control Daphnia pulex from the ENSR laboratory and may reflect contamination or a problem with analytical technique.

While these comparisons casts some doubt on the reliability of the measurements, the fish exposed to the WSP/2X treatment accumulated higher concentrations of As, Cu, and Pb than the fish exposed to the CON/0X diet. The concentrations of Cd, Cu, and Pb were higher in the fish fed the ENSR WSP/2X diet than in fish fed the CFI/2X or WS/1X (Woodward et al. 1994, 1995).

Conclusion. The use of metals from a naturally contaminated source and the control of dietary variables other than metals supports previous documentation that dietary metals are harmful to fish. The results observed by ENSR (1995) are very similar to effects observed by Woodward et al. (1994, 1995). However, the amount of difference in species composition between control and test diets was less in the ENSR (1995) study than in the Woodward et al. studies (1994, 1995). We do not believe that differences in species composition between control and test diets affected growth rate of fish in the Woodward et al. studies (1994, 1995), and the ENSR (1995) results adds support to that conclusion. The ENSR results represent additional information that is important in understanding the role of metals contaminated food-chains to early life stage fish in the Clark Fork River. These results add to a mounting body of evidence that food-chain invertebrates accumulate metals under natural conditions which are detrimental to age-0 trout in the Clark Fork River. These results with the

appropriate interpretation and analysis as we have provided herein, should be published in the primary literature to further our understanding of this important process.

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APPENDIX A. The following describe the methods and results for our analysis of the summary data provided in Table 3-6, page 3-10 of the document ENSR Number 0480-272 (ENSR 1995). We conducted standard two-sample t-tests with a Bonferroni correction for the 15 possible comparisons of the Control/Clark Fork and 0x,1x,2x factor combinations. Table A.1 summarizes the results. Table A.2 contains the test statistics and p-values for the individual comparisons. Significance of each comparison was indicated at the very conservative 0.05/15=0.00333 level.

Table A.1. Mean weight, length, condition factor and associated results of the Bonferroni t-tests for rainbow trout after 46 days of exposure. Means with the same letter are not significantly different (pairwise comparisons were tested at the conservative  $\alpha=0.05/15=0.00333$  level).

treatment	weight (g)		length(mm)			condition factor		
	mean	S.D. <sup>a</sup>	MCP <sup>b</sup> results	mean	S.D.	MCP results	mean	S.D.
control/0x	0.848	0.042	a	44.1	0.8	a	0.976	0.01 <sub>3</sub>
control/1x	0.866	0.015	a	44.1	0.2	a	0.997	0.01 <sub>7</sub>
control/2x	0.819	0.096	a b	43.8	1.0	a	0.946	0.04 <sub>0</sub>
cfr/0x	0.625	0.026	b c	40.1	0.7	b	0.952	0.02 <sub>5</sub>
cfr/1x	0.643	0.022	b c	40.9	0.2	b	0.937	0.03 <sub>9</sub>
cfr/2x	0.564	0.026	c	39.1	1.1	b	0.929	0.03 <sub>6</sub>

<sup>a</sup> standard deviation

<sup>b</sup> multiple comparison procedure (Bonferroni t-tests)

Appendix A2. Tables of p-values for t-tests for all possible pairwise comparisons of the Rainbow Trout weight, length and condition factor.

Comparison	weight		length		condition factor	
	t-statistic	p-value	t-statistic	p-value	t-statistic	p-value
Cont/0x vs. Cont/1x	0.8072	0.4504	0.0000	1.0000	1.9625	0.0974
Cont/0x vs. Cont/2x	0.5535	0.5999	0.4685	0.6560	1.4266	0.2036
Cont/0x vs. CFR/0x	9.0290	0.0001	7.5258	0.0003	1.7035	0.1394
Cont/0x vs. CFR/1x	8.6474	0.0001	7.7611	0.0002	1.8974	0.1066
Cont/0x vs. CFR/2x	11.4988	0.0001	7.3522	0.0003	2.4559	0.0494
Cont/1x vs. Cont/2x	0.9674	0.3707	0.5884	0.5778	2.3468	0.0573
Cont/1x vs. CFR/0x	16.0578	0.0001	10.9889	0.0003	2.9769	0.0247
Cont/1x vs. CFR/1x	16.7499	0.0001	22.6274	0.0001	2.8206	0.0303
Cont/1x vs. CFR/2x	20.1222	0.0001	8.9443	0.0001	3.4161	0.0142
Cont/2x vs. CFR/0x	3.9011	0.0080	6.0623	0.0009	0.2544	0.8077
Cont/2x vs. CFR/1x	3.5740	0.0117	5.6874	0.0013	0.3222	0.7582
Cont/2x vs. CFR/2x	5.1278	0.0022	6.3231	0.0007	0.6318	0.5508
CFR/0x vs. CFR/1x	1.0570	0.3312	2.1978	0.0703	0.6476	0.5412
CFR/0x vs. CFR/2x	3.3180	0.0160	1.5339	0.1759	1.0495	0.3344
CFR/1x vs. CFR/2x	4.6390	0.0035	3.2199	0.0181	0.3015	0.7732

## APPENDIX B

The reference diet/0x water treatment combination was excluded from analysis. With this treatment combination excluded, the design reduces to a two-factor analysis of variance. Levene's test was used to test for homogeneity of within treatment variances (6 variances, combinations of 2 diets and 3 metal concentrations in water). For all three variables, the homogeneity of variances assumption could not be rejected ( $\alpha=0.01$ ). Standard two-factor (diet and water) analysis of variance was conducted because the assumption of equal variances was met. Tukey's method for multiple comparisons was used to test for significant differences between levels of the main effects, since the interactions for all three variables were not significant.

### Length

The equality of variance assumption was not rejected ( $p=0.3213$ ). The interaction between diet and water was not significant ( $p=.1660$ ). The main effects of diet ( $p=0.0001$ ) and water ( $p=0.0429$ ) were both significant. Table 1 contains the results of Tukey's multiple comparison procedure.

### Weight

The equality of variance assumption was not rejected ( $p=0.0753$ ). The interaction between diet and water was not significant ( $p=.7402$ ). The main effects of diet ( $p=0.0001$ ) and water ( $p=0.0377$ ) were both significant. Table 2 contains the results of Tukey's multiple comparison procedure.

### Condition Factor

The means for each treatment are slightly different than those reported in Table 3-6 of the ENSR Number 0480-272. This is probably due to the fact that we calculated the condition factor for each tank using the mean weight and mean length for each tank, while ENSR probably calculated a condition factor for each fish and averaged these for the tank mean. The equality of variance assumption was not rejected ( $p=0.2196$ ). The interaction between diet and water was not significant ( $p=.2598$ ). The main effects of diet ( $p=0.0051$ ) was significant, while the main effect for water ( $p=0.2924$ ) was not significant. Table 3 contains the results of Tukey's multiple comparison procedure.

Table B.1. Means and results of Tukey's test for length. Means with the same letter are not significantly different( $\alpha=0.05$ ).

DIET			WATER		
LEVEL	MEAN	TUKEY GROUPING	LEVEL	MEAN	TUKEY GROUPING
CONTROL	44.00	A  B	1X	42.50	A A A  B A  B  B
CFR	40.05		0X	42.10	
			2X	41.48	

Table B.2. Means and results of Tukey's test for weight. Means with the same letter are not significantly different( $\alpha=0.05$ ).

DIET			WATER		
LEVEL	MEAN	TUKEY GROUPING	LEVEL	MEAN	TUKEY GROUPING
CONTROL	0.844	A  B	1X	0.754	A A A  B A  B  B
CFR	0.611		0X	0.736	
			2X	0.691	

Table B.3. Means and results of Tukey's test for condition factor. Means with the same letter are not significantly different( $\alpha=0.05$ ).

DIET			WATER		
LEVEL	MEAN	TUKEY GROUPING	LEVEL	MEAN	TUKEY GROUPING
CONTROL	0.990	A  B	1X	0.979	A A A  A  A A
CFR	0.950		0X	0.975	
			2X	0.956	







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**APPENDIX C**  
**CUTTHROAT TROUT AVOIDANCE OF INDIVIDUAL METALS UNDER**  
**CONDITIONS CHARACTERISTIC OF THE CLARK FORK RIVER AT**  
**ROCK CREEK, MONTANA**

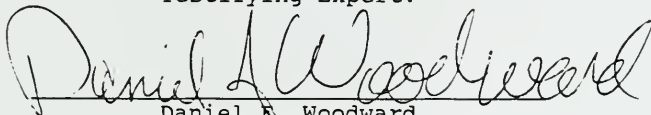


CUTTHROAT TROUT AVOIDANCE OF INDIVIDUAL METALS UNDER CONDITIONS  
CHARACTERISTIC OF THE CLARK FORK RIVER AT ROCK CREEK, MONTANA

Prepared by:

Daniel F. Woodward

Testifying Expert:

A handwritten signature in dark ink, reading "Daniel F. Woodward". The signature is written in a cursive style with a large, looping "D" and "W".

Daniel F. Woodward  
National Biological Service  
Midwest Science Center  
Jackson, Wyoming 83001

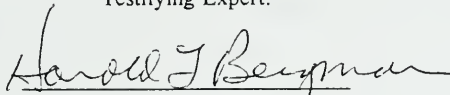
Date:

October 12, 1995



CUTTHROAT TROUT AVOIDANCE OF INDIVIDUAL METALS UNDER CONDITIONS  
CHARACTERISTIC OF THE CLARK FORK RIVER AT ROCK CREEK, MONTANA

Testifying Expert:

A handwritten signature in cursive script, reading "Harold L. Bergman". The signature is written in dark ink and is positioned above the printed name.

Harold L. Bergman, Ph.D.  
Department of Zoology and Physiology  
University of Wyoming  
Laramie, Wyoming 82071

Date:

October 12, 1995





Aquatic Resources Injury Report

Report of Additional Information

October 4, 1995

Cutthroat Trout Avoidance of Individual Metals Under Conditions  
Characteristic of the Clark Fork River at Rock Creek, Montana

Daniel F. Woodward

National Biological Service

Midwest Science Center

Jackson Field Station

Jackson, Wyoming 83001 USA

and

Harold L. Bergman

Department of Zoology and Physiology

University of Wyoming

Laramie, Wyoming 82071 USA

Metals contamination of the upper Clark Fork River basin in western Montana has been well documented (Johnson and Schmidt 1988, Luoma et al. 1989, Moore and Luoma 1990). The headwaters of the Clark Fork originate near the mining towns of Butte and Anaconda, which are known for their tailings deposits of copper (Cu), cadmium (Cd), lead (Pb), and zinc (Zn) (Miller 1973). Brown trout provide the main fishery in the upper Clark Fork River above Missoula, but the fish population is below carrying capacity of the river. Metal contamination is considered the primary cause of reduced fish populations (Johnson and Schmidt 1988, Chapman 1993). Copper and Zn occur in the river at concentrations frequently exceeding the water-quality criteria established in the United States to protect freshwater aquatic life, and Cd and Pb concentrations occasionally exceed the criteria for those metals (USEPA 1987, USGS 1989, Lambing 1991). The chronic criteria for these metals are hardness-dependent (varies with water hardness), but are at the following concentrations (ug/L) for the various metals at 100 mg/L hardness (as  $\text{CaCO}_3$ ): Cd, 1.1; Cu, 12; Pb, 3.2; and Zn, 110.

Water quality criteria were formulated by the United States Environmental Protection Agency to protect survival, growth, and reproduction of aquatic life; however, behavioral avoidance of contaminants may be an additional cause of reduced fish populations (Sprague 1968, Folmar 1976, Giattina et al. 1982, Giattina and Garton 1983, McNicol and Scherer 1991). Avoidance responses are particularly important because they occur at

concentrations lower than lethal thresholds (Little et al. 1993). Emigration resulting from avoidance of the metals may contribute to depressed fish populations and may be partially responsible for the low brown trout populations in the Clark Fork River.

Salmonids are particularly sensitive to metals, and they have been shown to avoid copper concentrations as low as 0.1  $\mu\text{g/L}$  (Folmar 1976). Introduction of Cu and Zn into a spawning tributary resulted in repulsion of ascending Atlantic salmon (Saunders and Sprague 1967). Prolonged exposure to metals has been shown to injure sensory tissue and impair or eliminate avoidance (Gardner and LaRoche 1973), allowing salmonids to be exposed to lethal concentrations.

Brown trout and rainbow trout have recently been shown to avoid low concentrations of metals mixtures mimicking conditions present in the Clark Fork River (Hansen 1993, Woodward et al. 1995). However, these studies raised new questions and additional avoidance experiments were conducted for the following purposes:

- #1 - to examine the role of individual metals and to determine which metals were most important in producing the avoidance response,
- #2 - to determine if metals concentrations, under conditions similar to those at the mouth of Rock Creek, would favor the movement of fish out of the Clark Fork River and into a clean water tributary, and
- #3 - to examine the avoidance response of a fish species

that is endemic to the tributaries of the Clark Fork Basin, westslope cutthroat trout (Oncorhynchus clarki lewisi).

## Methods

Experimental Fish. Due to difficulty in obtaining a disease free source of westslope cutthroat trout (Oncorhynchus clarki lewisi), Snake River Cutthroat trout (Oncorhynchus clarki spp.) were tested as a surrogate species. Fish were obtained from the Jackson National Fish Hatchery (Wyoming) as eggs, cultured at the Midwest Science Center (National Biological Service) Field Station, Jackson, Wyoming under the following conditions: temperature,  $10\pm 2^{\circ}\text{C}$ ; hardness, 160 mg/L as  $\text{CaCO}_3$ ; alkalinity, 150 mg/L as  $\text{CaCO}_3$ . Avoidance experiments were performed 3 to 5 months after hatching at size (gm/mm) range of 1.0/52 to 4/80. For each experiment, fish were acclimated to and maintained in the specified reference water for a minimum of four days before avoidance testing. Water temperature during testing was maintained at  $12\pm 0.4^{\circ}\text{C}$ .

Test procedure and analysis of water. Avoidance testing was performed in a plexiglass chamber similar to the counter-current design used in previous studies (Woodward et al. 1995). In brief, the avoidance chamber was designed so test or reference water could enter either end of the chamber ( $1,275\pm 100\text{mL}$  per min), flow towards, and exit out of the center. Following a 20 min acclimation period to reference water in both ends, the test water was allowed to enter a randomly selected end during a 30

min test period. The avoidance response was measured during the last 20 min of the test period. The response was recorded by video from which the following measurements were made: cumulative seconds on each side of chamber (time spent in test water versus reference water), percent time or cumulative frequency on each side of the chamber, number of trips into metals side of chamber, and trip time in seconds. The avoidance apparatus was enclosed in a structure to shield against external disturbances of movement, sound, and light. During each avoidance test, temperature differences between the reference and test end of the chamber was not more than 0.4°C.

During each experiment, water was sampled daily from test water and five or six times from the reference water to verify consistency of concentrations. Each water sample (150mL) was filtered using a Nalgene® 300 filter holder, transferred to a pre-cleaned, 125 ml I-Chem® polyethylene bottle, and preserved by addition of 1 ml Ultrex-II® nitric acid. The dissolved concentrations of Cd, Cu, and Pb in these samples were determined by graphite furnace atomic absorption spectrophotometry (Li et al. 1990). Zinc was determined by conventional flame atomic absorption spectrophotometry. Limits of detection were as follows (ug/L): Cd, 0.02-0.05; Cu, 0.62; Pb, 0.33-0.83; and Zn, 11. Blank samples were analyzed every 20 samples, and results for the blanks were always below detection limits. Quality control results for precision, accuracy, and percent recovery were all within  $\pm 10\%$  of ideal.

Avoidance of Individual Metals. We examined the role of individual metals to determine which metals were most important in producing the avoidance response. The general water characteristics were similar to those of Rock Creek at its confluence with the Clark Fork River (Table 1) (Lambing 1991). Characteristics were as follows: hardness, 50 mg/l; alkalinity, 50 mg/l; pH, 7.0 to 7.4; and conductivity, 150 uM/cm. Six waters were tested: Rock Creek -- with no metals added -- and five test waters in which Cd, Cu, Pb, and Zn were added to approximate the concentration of dissolved metals existing in the Clark Fork River at Turah Bridge (Tables 1 and 2). Of the five Clark Fork test waters, one had all four metals and four had only one metal: Clark Fork-Cd, Clark Fork-Cu, Clark Fork-Pb, and Clark Fork-Zn. Test waters were made up with reagent grade stocks:  $\text{CdCl}_2$ ,  $\text{CuCl}_2$ ,  $\text{PbCl}_2$ , and  $\text{ZnCl}_2$ . Rock Creek water was the reference water or the alternate choice for each test water. Rock Creek was selected to represent uncontaminated tributaries in the basin.

Statistical design and data analysis. Two avoidance chambers were used in each of the experiments which kept us from testing all treatments at the same time. A balanced incomplete block design was employed (Cochran & Cox 1957). The block (day) effects were not significant for any of the experiments and analysis of variance and Fisher's least significant difference test were performed on the data as in a completely randomized and balanced design ( $P \leq 0.05$ ). Percent time in test water was time in

test water expressed as a percentage (time in test water/total time (100)).

To meet the homogeneity of variance assumption for analysis of variance, total time, number of trips, and individual trip time were transformed by natural logs to equalize variances (Levene's Test,  $P \leq 0.05$ ). The equality of variances assumption was not met for the individual trip time variable. A closer look at this data indicated high variability in one treatment which probably accounted for rejection of the equal variances assumption. However, analysis of variance results continue to be fairly robust to unequal variances when designs are balanced and these results are reported (personal communication, W.P. Erickson, Western Ecosystems Technology, Inc., Cheyenne, Wyoming).

## Results

Measured metals concentrations. Concentrations of Cd and Pb measured in the test waters were usually within 10% of the nominal concentration (Table 2). Copper and Zn existed at measurable concentrations in background (measured concentration was greater than limits of detection when the nominal concentration was 0). Therefore, the measured Cu and Zn concentrations for all treatments were frequently higher than nominal. Background concentration for Cd and Pb were at the limits of detection. When the nominal was 0, the measurements for Cd and Pb were variable and could be reported as less than



the limits of detection.

Metals were not added to the Rock Creek water used in the avoidance experiments, and measured concentrations were near background and similar to field measurements made from March 1985 to October 1990 (Tables 1 and 2). The metals measured in the Clark Fork water in our experiments were also similar to the mean concentration reported for the Turah Bridge station, except for Zn. Following is a comparison of the means for the four metals of concern between Lambing's (1991) field measurements and measured concentrations (ug/L) in our Clark Fork water: Cd, <1 and 0.61; Cu, 7 and 7.1; Pb, 1 and 1.7; and Zn, 11 and 68. We had lower limits of detection for Cd, Cu, and Pb, but we had higher limits of detection and higher background for Zn.

Avoidance of Individual Metals. Cutthroat trout avoided water having metals concentrations similar to that of the Clark Fork River (at Turah Bridge) showing a significant preference for lower metals water that is characteristic of clean tributaries like Rock Creek (Table 3). Copper and Zinc were the metals responsible. Fish spent 6.3% of their time (76 of 1200 sec) in Clark Fork water containing Cd, Cu, Pb, and Zn versus 93.7% of their time in Rock Creek reference water. Time in Clark Fork water was significantly less than time spent in the Rock Creek test water with lower metals, 44% (523 of 1200 sec). When Clark Fork water contained only one of the four metals, cutthroat trout avoided those waters having either Cu or Zn, spending 7.7% (92 of 1200 sec) and 8.2% (98 of 1200 sec) of their time, respectively,

in those waters. However, fish did not avoid the Clark Fork water when Cd or Pb were the only metals present. Time spent in the Cd and Pb test waters was not significantly different from the Rock Creek water with no metals: Rock Creek, 44% (523 of 1200 sec); Rock Creek-Cd, 55% (657 of 1200 sec); and Rock Creek-Pb, 48% (570 of 1200 sec). Therefore, cutthroat trout are selective towards what they avoid, and Cu and Zn are the metals in the Clark Fork water which the fish are avoiding.

Mean trip time for cutthroat trout moving into Clark Fork water and Clark Fork water with Cu and Zn is significantly reduced to 4.1 sec or less, as compared to a 12 sec trip time for the Rock Creek water. There is a trend towards reduced number of trips into Clark Fork water and Clark Fork water with Cu and Zn, but this effect is not significant when compared to Rock Creek water.

## Discussion

Mean time in test water was the single most important measurement of avoidance and was a function of number of trips into the test water and length of time of each trip. The response of fish in the test waters could be compared to the 95% confidence interval for mean time in test water of those fish in the Rock Creek test water (Figure 1). For treatments with means falling inside the confidence interval, differences were not significant; conversely, means outside the confidence interval were significantly different.

The avoidance response is due to specific substances, Cu and Zn; and the avoidance response to Cu and Zn existing alone is similar to the avoidance response in a mixture of Cd, Cu, Pb, and Zn. The absence of cutthroat trout avoidance to Cd and Pb indicates the fish are responding to specific stimuli and not to a foreign material that is added to the test water. The absence of avoidance to Cd and Pb in our studies is also supported by the literature. The avoidance response of lake whitefish (Coregonus clupeaformis) to Cd was questionable in earlier studies (McNicol and Scherer (1991), but more recently considered non-existent (McNicol and Scherer 1993). Giattina and Garton (1983) found the avoidance threshold for rainbow trout to Pb was 26 ug/L, a concentration higher than those tested in this study.

Past studies at our laboratory have documented behavioral avoidance of metals under conditions simulating the upper Clark Fork River (Woodward et al. 1995, Hansen 1993). The avoidance response of rainbow trout and brown increased with increased concentration of metals. Hansen also found that rainbow trout can detect differences in two different metals concentrations and will prefer the lower metals water, even after 45d of acclimation to the higher metals water.

In these experiments, the cutthroat trout avoidance response to water simulating the Rock Creek and the Clark Fork River can be attributed to Cu. While fish avoided Zn also, Cu is the metal with the most elevated concentration when comparing the Clark Fork River with its tributaries (Lambing 1991). Mean dissolved

Cu was less than 3 ug/L over a 5-year sampling period (1985-1990) for the following tributaries: Little Blackfoot River, Flint Creek, Rock Creek, and Blackfoot River. Corresponding measurements for mean dissolved Cu in the Clark Fork River were 14 ug/L at Galen, 16 ug/L at Deer Lodge, and and 7 ug/L at Turah Bridge. Our studies suggest that fish encountering these differences in Cu concentrations would migrate out of the Clark Fork River and into the tributaries with lower Cu concentration.

Therefore, the concentrations and conditions where avoidance was demonstrated by cutthroat trout in our laboratory experiments are also present at the mouth of Rock Creek and other clean water tributaries in the Clark Fork basin. Fish populations in the Clark Fork River are dependent on recruitment from tributaries (Johnson and Schmidt 1988), and elevated metals -- copper in particular -- would discourage movement of fluvial stocks and reduce recruitment capability.

The results of our laboratory experiments along with the copper and fish monitoring data in the field provide a cause-and-effect relationship between elevated copper and a reduction in natural fish populations by behavioral avoidance. Behavioral avoidance of copper is a very sensitive response in cutthroat trout, and we believe this response plays a role in reducing fish populations and affecting movements of fish in the Clark Fork basin.

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Table 1. Statistical summary of water-quality data for periodic samples, March 1985 through October 1990 (Lambing 1991); N = 24-34, < = less than detection limits, -- = too low to calculate.

Parameter and unit of measure	Metals concentration, ug/L		
	Maximum	Minimum	Mean
Rock Creek near Clinton			
pH (standard units)	8.4	6.9	7.7
Temperature (°C)	18.0	0.0	8.6
Hardness (mg/L, CaCO <sub>3</sub> )	90	26	49
Alkalinity (mg/L, CaCO <sub>3</sub> )	82	22	45
Cadmium (dissolved, ug/L)	1	<1	--
Copper (dissolved, ug/L)	5	<1	--
Lead (dissolved, ug/L)	5	<1	--
Zinc (dissolved, ug/L)	15	<3	--
Clark Fork River at Turah Bridge			
pH (standard units)	8.7	7.4	8.0
Temperature (°C)	22	0.0	9.2
Hardness (mg/L, CaCO <sub>3</sub> )	210	67	142
Alkalinity (mg/L, CaCO <sub>3</sub> )	147	52	101
Cadmium (dissolved, ug/L)	1	<1	--
Copper (dissolved, ug/L)	25	2	7
Lead (dissolved, ug/L)	7	<1	1
Zinc (dissolved, ug/L)	39	<3	11



Table 2. Nominal and measured concentration of metals in waters used to test the avoidance response of fish to individual metals. For each water location, first row of numbers is nominal concentration; second row is N and mean measured concentration (standard deviation) for each metal, nm = not measured.

Location	Metals concentration, ug/L			
	Cd	Cu	Pb	Zn
Rock Creek	0	0	0	0
10	0.05 (0.03)	1.3 (1.0)	0.70 (0.09)	20 (8.5)
Clark Fork	0.66	6.0	1.3	55
5	0.61 (0.03)	7.1 (0.71)	1.7 (0.74)	68 (4.8)
Clark Fork-Cd	0.66	0	0	0
5	0.58 (0.04)	nm	0.84 (0.26)	24 (17)
Clark Fork-Cu	0	6.0	0	0
5	0.07 (0.05)	7.4 (1.0)	0.84 (0.25)	22 (5.5)
Clark Fork-Pb	0	0	1.3	0
5	0.07 (0.07)	nm	1.3 (0.21)	41 (15)
Clark Fork-Zn	0	0	0	55
5	0.06 (0.05)	nm	0.72 (0.10)	66 (6.7)

Table 3. Avoidance of metals at concentrations simulating conditions at the mouth of Rock Creek on the Clark Fork River. Metals were tested in a mixture and individually.

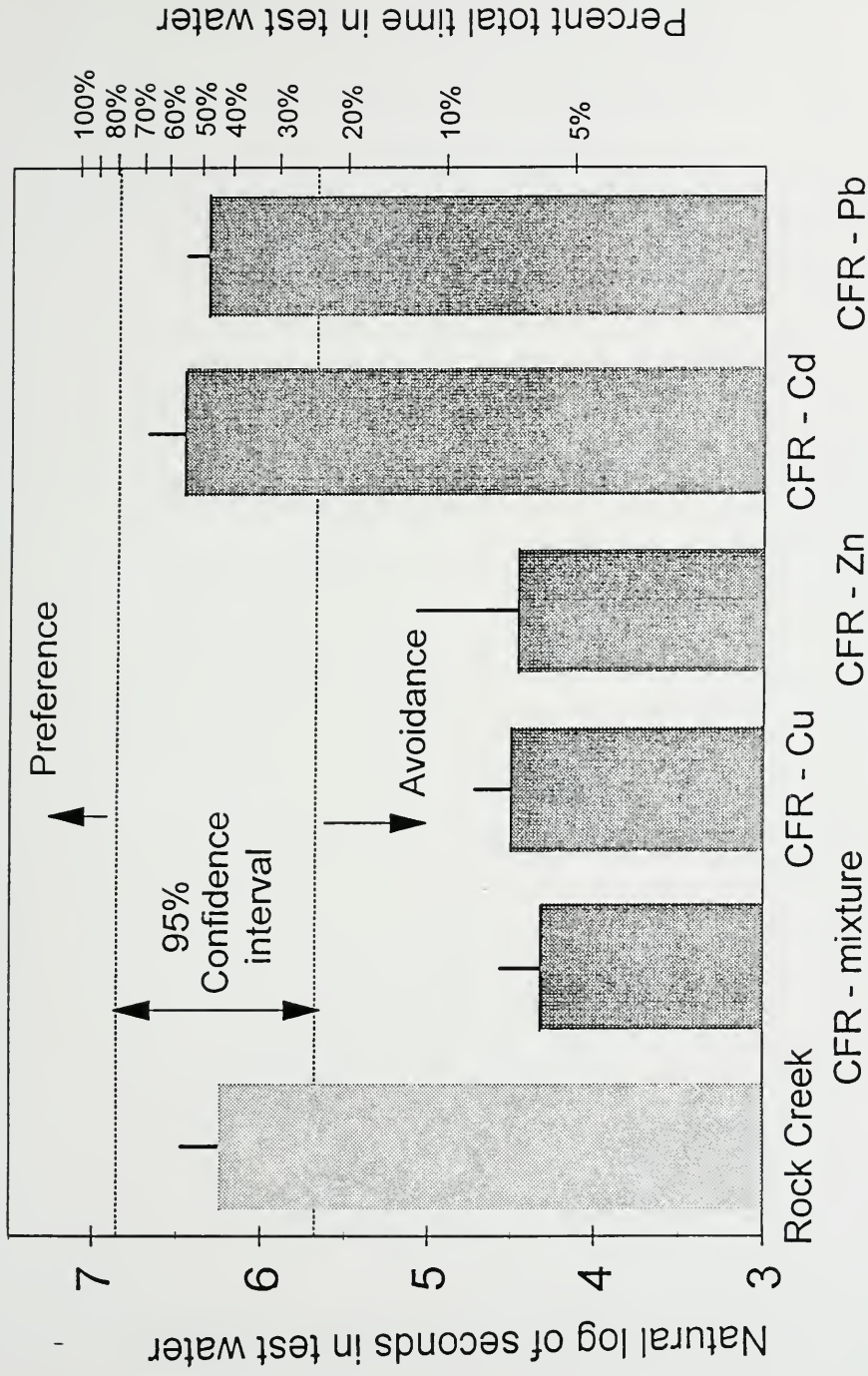
Location	Total time in test water (sec)	Percent time in test water	Number of trips into test water	Mean trip duration in test water (sec)
Rock Creek	523 (109)	44	50 (9.4)	12 (0.9)
Clark Fork	76 (18) <sup>a</sup>	6.3 <sup>a</sup>	35 (6.5)	2.2 (0.2) <sup>a</sup>
Clark Fork-Cu	92 (23) <sup>a</sup>	7.7 <sup>a</sup>	26 (11)	4.1 (0.6) <sup>a</sup>
Clark Fork-Zn	98 (50) <sup>a</sup>	8.2 <sup>a</sup>	29 (9.0)	3.2 (0.7) <sup>a</sup>
Clark Fork-Cd	657 (150)	55	46 (10)	19 (9.6)
Clark Fork-Pb	570 (81)	48	47 (8.4)	16 (4.8)

<sup>a</sup> Significantly different from Rock Creek reference water

(Fisher's least significant difference, ( $P \leq 0.05$ ))

### List of Figures

Figure 1. Percentage of time cutthroat trout spent in six test waters simulating metals concentrations at the mouth of Rock Creek on the Clark Fork River. Metals were tested in a mixture simulating the Clark Fork River (CFR) and individually (CFR-Cu, CFR-Zn, CFR-Cd, and CFR-Pb).



Test water

FIGURE 1.







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**APPENDIX D**  
**COMMENTS ON ARCO'S REPORTS CONCERNING THE STATE OF**  
**MONTANA'S INJURY ASSESSMENT FOR THE CLARK FORK RIVER -**  
**CHAPMAN**





**COMMENTS ON ARCO'S REPORTS  
CONCERNING THE STATE OF MONTANA'S  
INJURY ASSESSMENT FOR THE CLARK FORK RIVER**

**by**

**Donald W. Chapman, Ph.D.**

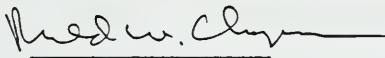


COMMENTS ON ARCO'S REPORTS  
CONCERNING THE STATE OF MONTANA'S  
INJURY ASSESSMENT FOR THE CLARK FORK RIVER

*Prepared by:*

Donald W. Chapman

*Testifying Expert:*



Donald W. Chapman, Ph.D.  
Don Chapman Consultants, Inc.  
3653 Rickenbacker, Suite 200  
Boise, ID. 83705

*Date:*

October 13, 1995



3653 Rickenbacker, Ste 200 • Boise, Idaho 83705  
(208) 383-3401 • FAX (208) 344-4861



**COMMENTS ON ARCO'S REPORTS  
CONCERNING THE STATE OF MONTANA'S  
INJURY ASSESSMENT FOR THE CLARK FORK RIVER**

by

**D. W. Chapman**

The State of Montana prepared reports on fishery damages caused by mining in the Clark Fork River (CFR). The Atlantic Richfield Company (ARCO) retained witnesses to prepare expert reports that criticize State reports. Below I rebut certain assertions by ARCO witnesses. I divide my rebuttal into three chapters; the first deals with classification, the second with channelization, and the final one treats water temperature in the CFR.

**Chapter I. Classification as a stratification tool  
in  
stream damage assessments**

This chapter discusses methods for selecting similar stream reaches in assessments of fishery losses. Such losses may occur as a result of chronic mine-related damage. Specifically, I address classification in the Clark Fork River (CFR) and potential reference or control sites chosen for their similarity to CFR stream sections. ARCO (Reiser 1995, p. C-6) criticizes use of classification to find stream sections similar to CFR sections.<sup>1</sup>

***I.A. What is stratification?***

The State of Montana used a system of stratification called "classification" to find stream sections similar to those in the CFR. Classification is a way of describing stream sections. It has many proponents. For example, Fausch et al. (1988) proposed that precision of predictive models in fisheries could be improved by stratifying lands into homogeneous ecoregions.

Stratification is a process of grouping similar units. It is used in statistical sampling to increase precision over that obtainable with simple random sampling

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<sup>1</sup> ARCO (Reiser 1995, p. C-6) states: "...the State's entire approach is based on the assumption that there is a direct relationship between the parameters included in its hierarchical classification scheme (ecoregion, geologic type, land type, valley bottom type and state type) and fish species composition and abundance. My review of the fisheries literature has not found any studies or research results which support this assumption..."

Levy and Lemeshow (1991). Levy and Lemeshow (1991, p. 105) note that the strategy for constructing strata has two steps: *"First, we determine the population parameter we are interested in estimating. Then we stratify the population with respect to another variable that is thought to be associated with the variable of interest. If our assumption of association is correct, this second step ensures that the strata are homogeneous with respect to the variable under consideration."*

An example of stratification in aquatic science would be to examine biological productivity in a large group of lakes. One might use fish production as the measure of biological richness. One could simply randomly sample from, say, 500 lakes scattered across the area of interest. Or one could stratify the lakes available for sampling by depth, surface area, and nutrient availability, expecting fish production to vary with size of the shallow areas available for light penetration, and with dissolved nutrients for plant production. Random sampling within the lake strata so-defined should produce data with much lower variance than would simple random sampling without stratification. Stratified random sampling should also be cheaper and more efficient.

We stratify our surroundings as we perceive them. We often use age, size, or behavior to stratify, noting: *"Teenagers have more energy," "Heavy cars use more gasoline than small cars,"* or *"Brown trout are harder to catch than cutthroat trout."* Political pollsters use stratification to select samples to determine the status of issues or politicians as elections approach.

### ***I.B. Support for stream classification as a stratification tool***

The evolution of classification for watersheds and streams is described in detail by McCullough (1988). Terrestrial classification systems developed from relationships between climate and soils, to response of vegetation to those factors. Climatic zonation has been recognized as correlated with vegetation distribution (Daubenmire 1956) and landform (Derbyshire 1976). In aquatic classification, fish production relates to geomorphology (Ziemer 1973; Swanston et al. 1977; Benda et al. 1992; Platts 1974).

Larsen and Omernik (1986) state: *"The physical character of a stream is controlled by the physical characteristics of its watershed, for example, land-surface form, geology, soil, and climate. Similarly, the stream's chemical character depends upon the interaction of precipitation falling on its catchment with these same terrestrial characteristics, modified by land use. The distribution and abundance of organisms are determined in part by the physical and chemical habitats created in these watersheds. To some extent their spatial patterns reflect spatial patterns in the physical-chemical character of the streams and watersheds. If streams depend on watersheds for their character, then stream systems can be classified according to mapped patterns of terrestrial characters."* Larsen and Omernik (1986) examined several case studies in Ohio, sampling fish assemblages relative to ecoregions. Differences in fish assemblages corresponded to ecoregion pattern.

Climate, geology, and vegetation set the stage on which the shaping of a stream channel takes place (Hunter 1991). Hunter (1991, p. 40) states: *"Climate dictates the amount and distribution of precipitation in a watershed, which ultimately affects the runoff collected by a river. The way precipitation shapes a landscape is determined by the geologic characteristics that dominate the drainage. The richness and texture of the soil - functions of climate and geology interacting with vegetation - influence both the amount of sediment that is washed off the landscape and into a stream ....."*

The United States was zoned by Omernik (1987) into 76 ecoregions, based on regional patterns in land surface form, soil, potential natural vegetation, and land use. Hughes et al. (1990) found that [1] fish communities demonstrate ecoregional patterns; [2] ecoregions that differ widely in landscape attributes support very different communities; [3] similar ecoregions support similar communities; and [4] within-region variation was less than among-region variation. CRAE (1992) concluded: *The use of reference sites within ecoregions thus appears to be a useful way of establishing criteria for restoration and recovery of RREs (riverine/riparian ecosystems).*

Hawkes et al. (1986) examined presence-absence data for 39 fish species from 410 streams sites in Kansas. The analysis confirmed ten ecologically meaningful fish assemblages, based on species associations. Mean annual runoff, mean annual growing season, and discharge appeared important, while mean width, mean depth, chloride concentration, water temperature, substrate type, gradient, and percent of pool habitat were less important. They noted that correspondence existed between fish ecoregions and the patterns of physiographic regions, river basins, geology, soil, and potential natural vegetation.

Frissell et al. (1986) state: *By viewing streams as hierarchically organized systems, the approach focuses on a small set of variables at each level that most determine system behaviors and capacities within the relevant spatiotemporal frame. Microscale patterns are constrained by macroscale geomorphic patterns.....Stream communities can be viewed as systems organized within this hierarchical habitat template."*

Classification is also supported by work on correspondence between classification elements and spatial patterns in streams (Whittier et al. 1988). Whittier et al. (1988) note: *"....Omernik's (1987) map of ecoregions of the conterminous United States can serve as a geographic framework for classifying streams. As a whole, streams within an ecoregion tend to be like other streams in that region and unlike streams in other regions. Regions with distinctly different land characteristics have streams that are more distinct; transitional regions have transitional characteristics."* Fish populations are affected both by the lands around them and by the hydrology of their drainage basin (Lotspeich and Platts 1982). Nelson et al. (1992) support this assertion with work on cutthroat trout. They found that trout distributions at specific sites were clearly related to geologic district and landtype association. They noted that location of study areas within discrete land classification taxa provides a great deal of preliminary information



about trout fishery potential. They state: *The results of this study illustrate the application of land classification to studies of trout distribution and habitat characteristics. We have emphasized not only the importance of rather broad taxonomic units (e.g., ecoregions), but also the utility of looking at smaller taxa.*" Lanka et al. (1987) report evidence that stream habitat is a function of geologic processes within the drainage basin. Benda et al. (1992) found that morphology and distribution of salmonid habitats in the Stillaguamish River basin (Washington) are related to geomorphology of a river basin at three spatial scales, including reach ( $10^2$ - $10^3$  m<sup>2</sup>), subbasin (2-26 km<sup>2</sup>), and watershed (240 km<sup>2</sup>).

Nawa et al. (1991) studied field survey data in 16 coastal Oregon watersheds in 1987-89. They found that many effects of land use were masked when all streams were considered, but became clear when data were stratified by segment type. They note: *"Segment classification provides a useful tool for reducing geomorphically-induced variation in habitat data, significantly increasing the power of standard survey and monitoring techniques."*

Platts (1974) reported that streams within a 397 mi<sup>2</sup> area in the upper South Fork Salmon River had distinguishing structural features that resulted from geomorphic processes. Streams that drained similar lands had been formed by similar processes, and therefore were relatively uniform in structure. Harris (1988) examined 10 streams in California, classified them into six geomorphic valley types and sampled them for environmental and riparian vegetation conditions. Significant associations between geomorphic valley types and riparian communities were found.

Rohm et al. (1987) evaluated the efficacy of a regional stream classification system based on the principle that streams reflect the character of the lands that they drain. Working with fish, physical habitat, and water quality data sets, they rejected the null hypothesis that classification did not reflect fundamental differences among the streams. They concluded that a regional stream classification system provides a sound rationale for spatially extrapolating results to other streams in the region, stating: *"The identification of regions containing streams with similar character can aid in selecting reference streams for impact assessments or in selecting monitoring sites from which results may be extrapolated regionally."*

Classification at the stream level could be used to infer species distribution, fish production potential, or aquatic primary production potential. A classification system could also be used to evaluate the watersheds that would provide largest returns on investment in stream habitat enhancement (McCullough 1988). Ziemer (1973) correlated drainage system geometry and salmon production in streams in Prince William Sound.

### ***1.C. Is classification at odds with the river continuum concept?***

The river continuum concept holds that from headwaters to mouth, physical variables within a river system present a continuous gradient of physical conditions

(Vannote et al. 1980). Some treatments of the river continuum concept assume homogeneity within a stream section of given order (order correlated with number of feeder watersheds upstream at a given stream point) (e.g., Minshall et al. 1983). However, Hughes and Gammon (1987) found that stream order was an inappropriate predictor of the diversity or composition of Willamette River fish assemblages.

Frissell et al. (1986) note that stream habitats and their communities vary and are spatially diverse within stream "segments." They define a segment as a portion of a stream system flowing through a single bedrock type, and bounded by tributary junctions or major waterfalls. A stream segment would compare approximately to the stream reaches defined by the State of Montana. Within reaches, the State additionally stratified or classified by "state." This class helped remove variability and diversity caused by stream channel changes that result from human activities along and within reaches.

The classification system used by the state is not contrary to the river continuum concept. Rather, it classifies to similar reach characteristics within stream sections, including many culturally-caused discontinuities.

***I.D. Does classification lead to similarity of CFR and reference areas with respect to land uses?***

Jensen (1995) examined land uses in the CFR and reference basins by determining land uses in the ecoregions in which the test and reference were located. He found considerable similarity in test and reference comparisons for the CFR and Rock Creek pairings (Northern and Middle Rocky Mountains Ecoregions), with range use 6% higher for the CFR and forest use 5.6% higher for Rock Creek. For test and reference comparisons in the CFR and Flint Creek, Big Hole River, Beaverhead, and Ruby River basins (Montana Valley and Foothill Prairie Ecoregions), Jensen (1995) reported considerable similarity in land uses. The Ruby River basin had higher agricultural, and the Beaverhead basin higher range proportions, than the CFR. He noted that higher proportions of agricultural and range uses in reference streams would tend to increase negative effects on stream resources there.

Flint Creek and the Big Hole River basins have greater proportions of irrigated land than the CFR basin. Irrigated proportions of Flint Creek, Rock Creek, and the Big Hole River are within 10% of the irrigated proportions of the CFR.

Jensen (1995) characterizes Omernik (1987) as stating that *"....the primary function of the ecoregion map is to provide a framework to: [1] compare similarities and differences of land/water relationships; [2] establish water quality standards that are in tune with regional patterns of tolerance and resilience to human impacts; [3] locate monitoring, demonstration, or reference sites; [4] extrapolate from existing, site-specific studies; and [5] predict the effects of changes in land use and pollution controls. Use of ecoregions as the broadest level of the hierarchical classification is appropriate."*

I conclude that classification resulted in pairings of streams that drain watersheds with similar land uses and irrigation patterns.

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## Chapter II. Channelization in the Clark Fork River

In materials prepared for ARCO, Reiser (1995) and HabiTech (1995) criticized the State of Montana for not considering extensive channelization in the Clark Fork River (CFR) as a cause of reduced trout populations there, and for ignoring upstream/downstream effects of channelization. This memorandum responds to ARCO assertions. It points out that all channelization is not equal in habitat effect, that the CFR is not channelized radically, that effects of channelization upstream and downstream cannot be assumed, and demonstrates quantitatively that our comparison of channelized CFR sections with straight reference sections in other streams is valid. I reject the contention of ARCO that channelization in the CFR invalidates the conclusion of the State of Montana that mining effects have reduced trout populations in the CFR. I reject the contention that channelized states in the CFR cannot be compared with straight sections in reference streams.

### II.A. *Is all channelization equal?*

It is important to examine what ARCO means by the term "channelized." ARCO (Reiser 1995, p. C-7) contends that the State paired one of the best state types (straight) in reference streams with one of the worst (channelized). ARCO provides Figure 6-12, p. D6-22 in Reiser (1995), stating that *"The lower photo illustrates a section of the Clark Fork River which has been channelized along the I-90 corridor; note the absence of any riparian vegetation and lack of instream habitat (boulders, logs); over 50 miles of the CFR have been similarly channelized."* This color photo and the associated text grossly misleads the reader, implying that 50 miles of the CFR look like this. On p. D6-26 (Reiser 1995), ARCO refers to Figure 6-12, contending that about 50 miles of the CFR have been channelized: *"Most of this channelization has occurred downstream of Deer Lodge where there are extensive reaches of the river which are straightened and lined with riprap (large angular boulders used to stabilize banks and prevent erosion)."* If one examines the foregoing ARCO statements, one would literally conclude that 50 miles of the CFR lack boulders as instream habitat, but that in most of those miles the bank is lined with riprap consisting of large angular boulders. These two contentions contradict each other. Both conditions cannot exist at the same time.

ARCO data demonstrate that channelization has affected sinuosity of the CFR little in comparison to reference streams. ARCO (Reiser 1995, p. D6-26) states that *"Most of this (CFR) channelization has occurred downstream of Deer Lodge where there are extensive reaches of the river which are straightened ..."* A casual reader would conclude that the CFR is a straight channel over much of its length. Yet ARCO (HabiTech 1995, p. B-70) shows sinuosities in all CFR reaches equal to or greater than in the Big Hole River and Rock Creek. Sinuosity of 1.47 in

CF3 (Gold Creek to Flint Creek), below Deer Lodge, exceeds that of reference streams Flint Creek, Rock Creek, and the Big Hole River. How can these sinuositities comport with ARCO's contention (Reiser 1995, p. D6-26)? The answer is that the Clark Fork meanders within the floodplain even though the latter is not as wide as it once was before construction of railroad and highway beds. Another part of the answer is that all channelization is not equal.

## ***II.B. Is riprap all bad?***

ARCO (HabiTech 1995, p. B-43) flatly states that *"The riprapping and diking commonly associated with channel straightening along highways and railroad lines destroys the natural streamside vegetation along the river corridor, thus directly reducing cover ....."* Yet Hunt and Graham (1972), evaluating riprap placement in the CFR, noted: *"The random placement of riprap containing a high percentage of pieces with volumes greater than one cubic yard each will provide bank stability and create a diversity of habitat for aquatic life. Riprap should be placed as steep as practicable within the slope stability limitations of the bank. The toe of the riprap should extend out into the stream to a depth below the design bed grade to prevent scour holes from undercutting the riprap."* Apparently not all riprap is equal.

HabiTech (1995) cites Bianchi and Marcoux (1975), noting that they pointed out the reduced cover value of riprap in comparison to natural streambanks. I can find no reference in Bianchi and Marcoux (1975) to the reduced cover value of riprap. Those authors tabulate information from Peterson (1974), which compared estimated number and pounds of trout in a bulldozed stream section with the same variates in a natural and in a riprapped stream section. The size of the riprap and details of habitat in the various sections were not provided in either Peterson (1974) or in Bianchi and Marcoux (1975). Furthermore, one has no way to determine, either from the information in the latter two papers or from HabiTech (1995) whether the modified and natural stream reaches would fall into similar strata if classified, hence would be similar and suitable for the pairings offered in Peterson (1974). No information is provided to permit a microhabitat comparison. In summary, ARCO (HabiTech 1995) relies on comparisons of reaches within given streams (especially the Ruby River) for evidence that riprapping is always bad. It provides no evidence that the reaches were sufficiently similar to validate the comparisons.

The HabiTech (1995, p. B-43) stated aversion to riprapping contradicts Wesche (1985, p. 155). The latter states: *"Not only does riprap provide fish cover and macroinvertebrate habitat, bank erosion can also be slowed or stopped, thereby allowing recovery of natural vegetation."* Again, all riprap is not equal.

Does riprap reduce trout populations? The answer to that question should be constrained by site-specific conditions. Response of trout to riprap will depend upon particle size and placement. Large riprap can benefit trout populations. CRAE (1992) notes increases in numbers of brown trout over 6 inches by 35% and

average number over 10 inches by 66% where banks of Willow Creek, Wisconsin, were riprapped. Riprap was tabulated by CRAE (1992) along with bank covers, current deflectors, and brush bundles, as habitat restoration tools. CRAE (1992) shows Rosgen channel types B-1-1, B2, C1, C1-1, C2, B3, B4, B5, and C3 to C5 as amenable to good to excellent benefits from bank-placed boulder (riprap). These channel types are found extensively in the CFR.

Where large riprap particles have adequate water depths around them, they can provide fish cover (Hunt and Graham 1972; Hunter 1991). Boulder riprap dislodged into the River Camowen in Northern Ireland diversified instream morphology (Kennedy et al. 1983). Burkhard (1967) examined two sections of Tomichi Creek in Colorado, before and after highway construction channelized one of the sections. Comparing sections over four years, he found no significant changes (caused by channelization) in brown trout numbers, mean length of age groups, or length-frequencies. He also found no channelization-caused differences in macroinvertebrate populations. Burkhard (1967) stated: *"The large rocks lining both sides and the bottom provide more cover for fish and probably provide more total area for bottom organisms."*

Brookes (1988) notes: *"Experience with enhancement techniques, particularly in North America, has revealed the advantages of riprap, composed of natural rock or quarry stone, as opposed to more conventional stabilization methods....Vegetation can become established between the stones, particularly where a soil cover has been applied. It may also provide a stable substrate for benthic invertebrates and the weathering of stones may produce gravel beds suitable for fish spawning.....Increased primary productivity and abundance of invertebrates have been associated with riprap."*

Not all riprap benefits trout populations. Small riprap does not provide interstices and velocity diversity. Neither is it as stable as large particles. However, in stream reaches on which large woody vegetation is not and cannot be established, large-particle riprap can provide cover that would otherwise not exist. As long as water depth in winter permits, large-particle interstices can provide winter trout hiding places. Wesche (1980) notes that trout cover can generally be described as object-oriented (i.e., rubble, boulders, undercut banks, logs, vegetation), having a water depth of at least 0.5 feet and a water velocity at the point of cover occupation (point velocity) of less than 0.5 ft/sec. Large riprap elements can fit this definition.

## ***II.C. Does channelization degrade habitat upstream and downstream?***

ARCO consultant HabiTech (1995) states: *"Channelization can also result in fish habitat degradation both downstream and upstream from the altered stream reach."* In support, HabiTech (1995) cites Simpson et al. (1982) and Brookes (1988). In a paragraph on downstream effects, HabiTech uses "can impact," "can affect," "can result," "a possible shift." No documentation of downstream effects is offered for the CFR. In discussing effects upstream from channelized areas,



HabiTech (1995) admits they have been studied even less than downstream effects, and uses "possible upstream effects," "authors....speculate," "may result," "potential impacts," and "fish productivity in upstream river reaches can be negatively impacted by downstream channelization" [conditional bolding mine]. Close reading of Simpson et al. (1982) reveals considerable speculation but no empirical evidence applicable to conditions in either the CFR or reference streams. "May," "could," and "can" substitute for evidence.

Brookes (1988), the other passing cite offered by HabiTech (1995), concentrates on downstream effects of channelization, including flooding that results from reduced retentive capacity of channelized river reaches. However, Brookes (1988) notes that "*Channel improvement works may only have a local effect on water velocities....*" He also mentions increased scour and sediment transport that can result downstream from channelized segments that increase river velocity, using examples from the United Kingdom. Brookes (1988) rightly notes that construction activities associated with channelization can increase turbidity, sediment transport, and alter water quality. There is no question that channelization *can* have physical and ecological effects downstream from the altered channel segments. However, neither Simpson et al. (1982) nor Brookes (1988) offer empirical information useful in determining the degree to which channelization in segments of the CFR may have affected downstream stream morphology, ecology, and fish abundance.

In supporting its contention that trout populations are reduced by channelization, HabiTech (1995) cites various studies that have compared fish abundance in channelized and unchannelized reaches. One cite, a personal communication from A. Whitney mentioned in Peters and Alvord (1963) points out that a 300 ft channelization of Flint Creek (one of the State's reference streams) reduced fish populations in the section. If one follows ARCO's line, one would contend that there must have been upstream and downstream effects from that (and other) channelization in Flint Creek, hence the State's reference baseline populations are smaller than they would be without those upstream and downstream effects. This would mean that the State estimate of baseline populations on reference areas in Flint Creek was conservatively low. This would carry to other reference streams as well, e.g., to the Bighole River, where Peters and Alvord (1963) report 31% of the stream was altered before 1963, and in the Ruby River (HabiTech 1995).

Most workers discount upstream/downstream effects of channelization, for most evaluations of the effect of channelization compare channelized sections with unchannelized sections of the same stream. HabiTech (1995) appears not to believe that upstream and downstream effects of channelization affect fish populations, for when it compared alterations in reference streams with the CFR, it only reported percentage alteration *in reaches that contained reference sites*, ignoring alterations in the remainder of the river. For example, HabiTech (1995) examined 54.2 miles of the Big Hole River and reported 5% of it as altered. Peters and Alvord (1963) examined 147.6 miles of the Big Hole River, finding 46.1 altered

miles, or 31% alteration (by 1963). If HabiTech (1995) believed that upstream/downstream effects were important, it would more appropriately have examined all of the Big Hole River (and other reference streams) for alteration.

My purpose here is not to deny that channelization can cause reductions in habitat quality and fish abundance. Certainly it can. Rather, I point out that [1] evidence is lacking that channelization in the CFR affects fish populations upstream and downstream from the altered stream sections; and [2] extensive stream alterations have occurred in reference streams; and [3] the classification system that we used reasonably compared channelized CFR sections with straight sections of reference streams.

It is instructive to examine effects on trout populations where large reaches of a given stream were channelized. Elser (1968) examined altered (channelization affected one-third of the stream length) and unaltered sections of Prickly Pear Creek in Montana. He reported a total stream trout population of 20,400 (fish longer than 4.0 inches), and a total loss due to channel alterations of one-third of the stream length as about 4,700 trout; this would represent a loss of about 19% of potential standing stock of fish.

Peters and Alvord (1963) tabulate amounts of channel alteration in the Big Hole River. They report 4.4 miles of channel relocation and loss of 12.9 miles of natural stream channel, 11 miles of riprap at 107 sites, 17 miles of diking in 219 sites, and a total of 46.1 miles of altered stream (395 instances in 148 stream miles). They calculate that 31% of the Big Hole River was altered by 1963. This can be compared to Jensen's (1995) estimate that about 33% of the CFR has been altered. This estimate for the CFR is considerably lower than the 48% estimated by HabiTech (1995) or the "50% of its (CFR) length between Missoula and Warm Springs" estimate of Reiser (1995, p. C-26).

#### ***II.D. Can straight reference sections be compared to CFR channelized sites?***

ARCO (HabiTech 1995, p. B-38) takes issue with the State's use of straight reference reaches as comparable with channelized sites. HabiTech notes: "*While an altered, straightened channel may have a sinuosity and channel pattern similar to a natural straight reach, the substrate, water column and streambank conditions may differ substantially.*" It supports this observation with no empirical measurements, and with general observations from various authors (see HabiTech 1995, p. B-43).

Does classification permit us to "control" for stream channelization? Asked another way, do straight stream sections properly compare with channelized CFR sections? Instead of using general statements and literature not directly appropriate for the CFR, we compared quantitative microhabitat characteristics of paired channelized sites in the CFR and straight study sites in reference streams (Table II.1). In Table II.1 we included data for all channelized CFR sites v. all straight reference sites.

Table II.1. Comparison of microhabitat statistics in CFR and reference sites. Included are results of PHABSIM modeling and microhabitat statistics not treated in that modeling. Thus, no statistics dealing with area of stream or area of substrate are included. Data extracted from Appendices C2, C5, C7, C10, C11, and C13 of the State Assessment of Injury to Fish Populations: Clark Fork River NPL sites, Montana, dated January 1995.

Characteristic	Test CFR	Reference	Significance	Favors reference?
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**State 4/5, Reach 1**

Pool rating	4.43	0.93	yes	no
Mean depth, ft	4.67	1.68	yes	no
Thalweg depth, ft	7.88	2.86	yes	no
Canopy cover %	2.03	0.2	no	
Woody debris %	0	0.73	no	
Bank angle, deg	146	155	yes	no
Veg. overhang, in	1.03	0	yes	no
Bank cover, %	90	90	no	
Bank alteration, %	0	0	no	
Bank undercut in	0	0.23	no	
WUA m <sup>2</sup> /100 m site, brown adult	492	439	n/a	no
WUA m <sup>2</sup> /100 m site, rainbow adult	1661	1866	n/a	yes

**State 4, Reach 2**

Pool rating	2.33	0.27	no	
Mean depth, ft	1.35	1.01	yes	no
Thalweg depth, ft	2.48	2.16	yes	no
Canopy cover %	0	2.03	no	
Woody debris %	0	0.32	no	
Bank angle, deg	167	152	yes	yes
Veg. overhang, in	0	1.38	no	
Bank cover, %	90	90	no	

Bank alteration, %	0	0	no	
Bank undercut in	0	0	no	
WUA m <sup>2</sup> /100 m site, brown adult	462	772	n/a	yes
WUA m <sup>2</sup> /100 m site, rainbow adult	937	1490	n/a	yes

#### State 4, Reach 3.

Pool rating	0	0	no	
Mean depth, ft	0.87	0.74	yes	no
Thalweg depth, ft	1.66	1.73	no	
Canopy cover %	0.4	0.43	no	
Woody debris %	0	0	no	
Bank angle, deg	158	172	yes	no
Veg. overhang, in	0.12	0.35	no	
Bank cover, %	87	90	no	
Bank alteration, %	4.25	0	no	
Bank undercut in	0.45	0	no	
WUA m <sup>2</sup> /100 m site, brown adult	1116	557	n/a	no
WUA m <sup>2</sup> /100 m site, rainbow adult	2701	2297	n/a	no

#### State 4, Reach 4.

Pool rating	0	0	no	
Mean depth, ft	1.07	0.74	yes	no
Thalweg depth, ft	1.89	1.73	no	
Canopy cover %	0	0.43	no	
Woody debris %	0	0	no	
Bank angle, deg	156	172	no	
Veg. overhang, in	0	0.35	no	
Bank cover, %	87.5	90	no	
Bank alteration, %	2.58	0	no	

Bank undercut in	0	0	no	
WUA m <sup>2</sup> /100 m site, brown adult	861	557	n/a	no
WUA m <sup>2</sup> /100 m site, rainbow adult	2126	2297	n/a	yes

#### State 4, Reach 5.

Pool rating	0.3	0	no	
Mean depth, ft	1.53	0.86	yes	no
Thalweg depth, ft	3.94	1.45	yes	no
Canopy cover %	1	0	no	
Woody debris %	0.1	0	no	
Bank angle, deg	134	175	yes	no
Veg. overhang, in	0.45	0	yes	no
Bank cover, %	88.3	90	no	
Bank alteration, %	2.9	0	yes	yes
Bank undercut in	0.03	0	no	
WUA m <sup>2</sup> /100 m site, brown adult	767	767	n/a	no
WUA m <sup>2</sup> /100 m site, rainbow adult	1688	3373	n/a	yes

#### State 4, Reach 6.

Pool rating	0	2.1	yes	yes
Mean depth, ft	0.59	0.9	yes	yes
Thalweg depth, ft	1.34	1.46	no	
Canopy cover %	4.4	9.43	no	
Woody debris %	0.35	3.2	no	
Bank angle, deg	68.2	146	yes	no
Veg. overhang, in	1.93	1.4	no	
Bank cover, %	89.2	90	no	
Bank alteration, %	0.58	0	no	
Bank undercut in	0.87	0.03	yes	no

WUA m <sup>2</sup> /100 m site, brown adult	744	224	n/a	no
WUA m <sup>2</sup> /100 m site, rainbow adult	948	378	n/a	no

In 16 of 20 instances in Table II.1 in which microhabitat features differed significantly between CFR and reference sites, the difference favored CFR sites. This means that habitat in the channelized site tended to have more favorable features than in the reference site. In 4 instances in 20, the difference favored the reference site.

We also compared weighted usable area (WUA) as estimated in PHABSIM modeling in the six CFR channelized sites compared with straight reference sites (Table II.1). In 5 of 6 sites, the CFR channelized areas contained more WUA per 100 m for brown trout adults. In 4 of 6 sites, the CFR channelized areas contained less WUA per 100 m for rainbow adults. However, in 2 of the 4 instances in which WUA for adult rainbow favored the reference area, the percentages favoring the reference site were only 8% and 12%. Thus, one should conclude that the characteristics of test and reference sites do not support the contention of HabiTech (1995, p. B-43) and Reiser (1995, p. C-7) that pairing of straight sites in reference streams with channelized ones in the CFR biases results in favor of the reference sites. The available data on habitat characteristics indicate that the channelized CFR sites contain better-quality habitat than the straight reference sites with which we compared them. It is important to point out here that adjustment of fish densities with PHABSIM data helps compensate for habitat differences between CFR and reference sites. Stalnaker (1979) used data of Wesche (1976) to correlate standing crop of brown trout with WUA in eight Wyoming streams. For the 19 data sets used, the correlation was high ( $r^2 = 0.81$ ). This result tends to support our adjustment of abundance estimates on the basis of WUA. However, the key finding in the foregoing exercise is that ARCO is incorrect when it assumes that straight reference sections (randomly selected) are superior with respect to habitat quality in comparison to CFR channelized sections (also randomly selected).

Since ARCO consultants also criticized use of "altered" reaches in the CFR, it is worthwhile to compare trout populations in the reaches they do not term "altered." Table II.2 contains reach/state data for unaltered CFR reaches and the accompanying reference reaches.



Table II.2. Numbers of all trout per ha in reach/states termed by HabiTech (1995) as not altered (see p. B-39 and B-40 of HabiTech 1995) in either CFR or reference sites. Number of adult brown trout per WUA shown in brackets. Four 100 m sites make up each average (see Appendix H1 in State Damage Report).

Reach	State	Number/ha test [adult brown/WUA]	Number/ha reference [adult brown/WUA]
1	3/5	81.9 [1.1]	148.1 [4.5]
2	3	29.9 [0.7]	240.8 [8.0]
4	4 (1 of 4 sites "altered", 3 not)	5.8 [1.2]	19.2 [3.2]
3	3	7.0 [3.5]	50.3 [10.5]
4	3 (1 of 4 sites "altered", 3 not)	11.6 [2.4]	41.2 [3.0]
6	2d	10.1 [3.0]	458.8 [182]
6	2u	2.1 [0]	794.1 [334]
Mean of all		36.1 [1.7]	250.4 [77.9]
Mean without reach 4 data		26.0 [1.7]	338.4 [107.8]

The information in Table II.2 indicates that unaltered sites in the CFR had very low numbers of trout relative to their paired control sites, which also were unaltered (designation "altered" as offered by HabiTech [1995, p. B-39 and B-40]).

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### Chapter III. Water temperature in the Clark Fork River

ARCO (Reiser 1995, p. D-6-17) contends that the Clark Fork River (CFR) is inhospitable for rainbow trout because of high water temperatures upstream from the mouth of Rock Creek. It also contends that elevated water temperatures in the CFR likely cause brown and rainbow trout avoidance, reduced production potential, reduced macroinvertebrate density, and force trout to rely on tributaries as temporary refuges in summer. I find that ARCO applied generic temperature information without considering site-specific data for Montana streams, failed to demonstrate that macroinvertebrate density was reduced by the temperature regime in the CFR, and incorrectly interpreted empirical evidence on brown trout avoidance of CFR temperatures.

#### *III.A. How warm is too warm for rainbow and brown trout?*

Hill et al. (1993) tabulated maximum, minimum, and optimum water temperatures defined by salmonid literature (Table 1). They then plotted a tolerance zone and an optimum zone for various species (e.g., zones plotted for rainbow and brown trout compared to maximum, minimum, and average temperatures of the CFR at Clinton). That information indicates that the optimum rearing temperature in the Clark Fork can be exceeded in July and August.

Hill et al. (1993) and ARCO rely on generalized literature thresholds. However, specific responses of fish can belie those generalizations. Trout acclimated to high temperatures can tolerate higher temperatures (Elliott 1981). For example, Lee and Rinne (1980) demonstrated a higher upper lethal temperature for fish acclimated to 20° C. than to 15° C. In the warm lower Madison River, rainbow trout have an upper lethal limit of about 28° C. (Goodman et al. 1983). Goodman et al. (1983) also showed that substantial populations of rainbow and brown trout live in the lower Madison and that high temperatures there did not impair growth. Temperatures in the lower Madison River are higher in summer than those reported for the CFR. Rainbow and brown trout in the Firehole River also cope with higher temperatures than those in the middle CFR (see Koch 1990). Grande and Andersen (1991) report a critical thermal maximum temperature  $LT_{50}$  (temperature giving 50% mortality) for 2-3-month-old salmonids. It varied from 26.2° C. to 27.9° C.

Hill et al. (1993) note: *"Most literature values for temperature indicate that brown trout can tolerate higher temperatures than rainbow trout; however, important exceptions can be found in work on specific growth rates by Elliott (1975) on brown trout and Hokanson [et.al.] (1977) on rainbow trout. Data from these papers indicate that rainbow trout grow better at high temperatures than do brown trout....as shown in the lower Madison River (Goodman et al. 1983), rainbow trout cope better with higher temperatures than do brown trout."*

Hokanson et al. (1977) showed that growth rate in rainbow trout remained

high until water temperatures reached 22° C. The highest constant temperature at which specific growth and mortality rates became equal (initial test biomass remained constant over 40 d) was 23° C. The highest temperature at which a rainbow trout population can be expected to maintain its weight for 40 days was estimated for fluctuating temperatures as 21° C. Their work also suggested that rainbow trout acclimate to some value between the mean and maximum daily temperature when they live in fluctuating temperatures.

The temperature range from 21 to 23° C. has been reported by various authors as the upper limit of rainbow trout distribution. Hokanson et al. (1977) suggest that the maximum mean weekly temperature of 19° C. as set by the NAS/NAE (1972) should be retracted to 17° C. where diel temperature variations exceed  $\pm 2^\circ \text{C}$ . Grande and Andersen (1991) report the highest temperatures at which rainbow and brown trout fry fed were 26.6° C. and 26.2° C., respectively. They noted that temperatures of 25° C. or higher were required to kill, or reduce the appetite of, brown and rainbow trout.

Reiser (1995) suggests that absence of rainbow trout fry in the Clark Fork River can be explained by high summer temperatures. However, rainbow trout reproduction and early rearing in Montana rivers is concentrated in tributaries.

While temperatures in portions of the Clark Fork River exceed the optimum for rainbow trout in parts of July and August, on average, they do not appear to be high enough to prohibit rainbow trout use of the river.

### ***III.B. Do whitefish prefer lower temperatures than trout?***

Bovee (1978) prepared suitability-of-use curves for mountain whitefish adults, juveniles, and fry. He shows maximum use probability at 60° F. (15.6° C.) with a range from near-freezing to about 75° F. (23.8° C.). He shows a higher upper limit for rainbow trout and brown trout adults and juveniles, and about the same upper limit for rainbow and brown trout fry as for whitefish. Bell (1986) shows the preferred range of temperature for whitefish as 54-62° F. (12.2-16.7° C.), in comparison to his optimum temperature for rainbow as 57° F. (13.9° C.). Piper et al. (1982), apparently not reviewed by Bell (1986), shows an optimum range for rainbow as 13-17° C., close to Bell's (1986) preferred range for whitefish adults.

Like other salmonids, whitefish have different seasonal preferences. Prespawning preference of whitefish was for 17.7° C.; the preference in spring was 16.3° C. (Ihnat and Bulkley 1984). No summer preferences were obtained, so the prespawning (fall) preference would be the closest available to summer preference. The latter may actually be higher than the prespawning preference. Higher acclimation temperature leads to higher temperature preferences. The highest acclimation temperature tested by Ihnat and Bulkley (1984) was 15° C.

From the foregoing, I conclude that the available evidence does not support the hypothesis that whitefish have a higher temperature tolerance than rainbow trout. In fact, the reverse appears more likely.

### III.C. Are macroinvertebrate densities reduced by high temperatures in the CFR?

Although ARCO (Reiser 1995, p. D6-17) indicates that high temperatures in the CFR reduce invertebrate densities, no evidence is presented in Reiser (1995) that supports such a conclusion. If summer high temperatures affect invertebrate densities, one should expect the fall populations of invertebrates to reflect that effect.

With respect to macroinvertebrate diversity, Reiser (1995) states, concerning October, 1988, invertebrate communities in the CFR between the Little Blackfoot River (LBR) and Milltown Dam: *"Invertebrate diversity was relatively high and stable at all stations between the LBR and Milltown Reservoir, indicating a general increase in the number of taxa present, and decreasing dominance of hydropsychid caddisflies, chironomids, and simuliids which were more abundant in the section of the CFR between Warm Springs Ponds and the LBR."* Hence one is led to the conclusion that high temperatures in the lower CFR must not have reduced taxa diversity.

Reiser (1995) notes similar general longitudinal trends in diversity and taxa richness from year to year (p. D5-45). He also notes that the longitudinal trends indicate progressive changes in organic matter concentrations and nutrient enrichment.

Reiser (1995, p. D6-20) states that *"The temperature regime in the CFR has also likely influenced the composition and density of aquatic invertebrates, which are important sources of food for trout. The reduced abundance of mayflies, and particularly stoneflies, in the river between Warm Springs Ponds and the Little Blackfoot River are indicative of higher temperature in this section of the river when compared to the section between the Little Blackfoot River and Milltown Dam."* Reiser notes that temperatures sometimes reached 24° C., exceeding *"..the 20° C. value which has been determined to eliminate many types of mayflies and stoneflies from rivers and streams."* He offers no citations on this point, but also does not discuss whether he believes it is occasional attainment of temperatures above 20° C., or attainment of average temperatures above these levels that may cause mayflies and stoneflies to abandon the reach.

Finally, Reiser (1995) notes that stoneflies are often reduced in abundance and diversity below tailwaters of dams (Warm Springs Ponds). His text does not tell the reader whether it is temperature or tailwater effects, or both, that may be responsible for reduced stonefly abundance. At other points in his text, he makes it clear that he believes tailwater effects are most important in influencing diversity, and does not mention temperature (see p. D5-18, D5-13): *"The influence of the pond outfall on the CFR invertebrates diminishes as the suspended materials are removed by filter-feeding insects and/or settle out, as is evidenced by the reduced relative importance of the filter-feeding insects at downstream stations."* (see Reiser [1995], p. D5-21).

I can find no evidence in Reiser (1995) or elsewhere to support the contention that high temperatures in the Clark Fork have depressed

macroinvertebrate community diversity or abundance.

### ***III.D. Do trout use tributaries as refugia from high temperatures?***

Trout in the CFR, like those in most streams, will use groundwater inflow, flows from cool tributaries, and tributaries themselves at various times. Kaya et al. (1977) offer a Firehole River example. Movements in the Firehole into Sentinel Creek, the only cold-water tributary on the lower Firehole River, did not occur until mean daily water temperatures in the Firehole River exceeded 24° C. or daily temperature maxima exceeded 25° C. Cherry et al. (1975) found that preferred temperature for rainbow trout increased with increasing acclimation temperature. For example, when fish were acclimated to 24, 21, and 18° C., the preferred temperatures were 22, 20.1, and 18.1° C., respectively. Upper and lower temperatures avoided by rainbow trout acclimated to 24° C., 21° C., and 18° C. were 25°\18° C. (high and low temperature avoided), 23°\16° C., and 20°\14° C., respectively. In other words, when rainbow trout were accustomed to high temperatures, higher temperatures were required to elicit avoidance in laboratory experiments. It is important to note that trout in natural stream channels have options for avoidance of high or low temperature during the daily temperature cycle. They can seek microhabitat locations in groundwater or bank seepage, or in unmixed eddies where high or low temperatures exist. McCauley et al. (1977) found no effect of acclimation temperature on preferred temperature. Differing experimental conditions may explain the difference between fish responses in laboratory environments used by McCauley et al. (1977) and Cherry et al. (1975).

R2 Resource Consultants (1995) (Appendix J to Reiser [1995]) offers length frequency data from Gold Creek in 1990 (see Figure III.1, from R2 Consultants' Figure 3b) as evidence of movement into Gold Creek from the CFR as adult brown trout avoid warm temperatures in the latter. The length-frequencies in summer (August 1990) are indeed very different from those in fall, and notable because of virtual absence of fish smaller than 150 mm. One must ask where these small fish were in summer.

Ancillary questions arise about R2 Consultants' Figure 3a. The length-frequency of brown trout in the main adjacent CFR in spring, 1990, shows a strong mode at about 320 mm, while the summer length-frequency mode in Gold Creek does not. In fact, the Gold Creek summer length-frequencies are bimodal at 160 and 250 mm. The CFR length frequencies in spring are bimodal at 260 and at least at 320 mm. Very few fish over 350 mm appear in the Gold Fork summer length-frequencies. Furthermore, the CFR in spring lacked an obvious presence of brown trout of 120-200 mm that makes up a major part of the Gold Creek summer histogram. Does this suggest that that component in summer was not from the CFR at all, but from Gold Creek?

An explanation other than avoidance responses arises for R2 Consultants' Figure 3b: *The summer length-frequencies in Gold Creek in 1990 consisted of fish produced in Gold Creek.* Small brown trout (ca. 75-120 mm) produced in upper



Gold Creek moved downstream to take up favorable rearing positions by fall, 1990, changing the shape of the Gold Creek histogram. I recalculated the histogram for fall, 1990, leaving out fish smaller than 150 mm, to determine if the shape of the histogram was similar to the summer shape. The calculation was approximate, as it was necessary to read percentages for summer length-frequencies from Figure 3b in ARCO's Appendix J, and apply them to the 285 fish in the fall, 1990 sample. I estimated that 68 fish were included in the group of fish larger than 150 mm. This comports with the 106 fish present in summer, 1990. Some fish in the size complement larger than 150 mm would suffer mortality before fall, and some would have moved into the Clark Fork, or at least out of the sampling area. Figure III.2, attached, has a configuration remarkably similar to the summer, 1990 length-frequency histogram shown by the solid black bars in R2 Consultants' Figure 3b. I also reduced Figure III.2 and placed it above R2 Consultants' Figure 3b (see my Figure III.3). Figure III.3 clearly shows close correspondence of length frequencies for brown trout in Gold Creek in summer and fall, 1990.

I conclude that small fish moved into the sampling area from upstream in Gold Creek between summer and fall sampling. I find no justification for the ARCO argument in R2 Consultants' Appendix J that brown trout entered Gold Creek to escape unfavorable conditions in the Clark Fork.

### Reach R7 Brown Trout Length Frequency

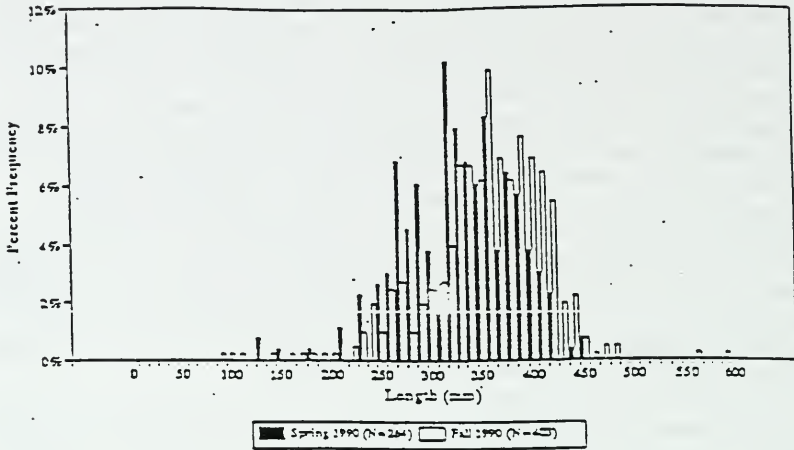


Figure 3a. Length frequency distributions for spring and fall, 1990 for brown trout populations in the mainstem CFR, in the vicinity of Gold Creek.

### Gold Creek Brown Trout Length Frequency

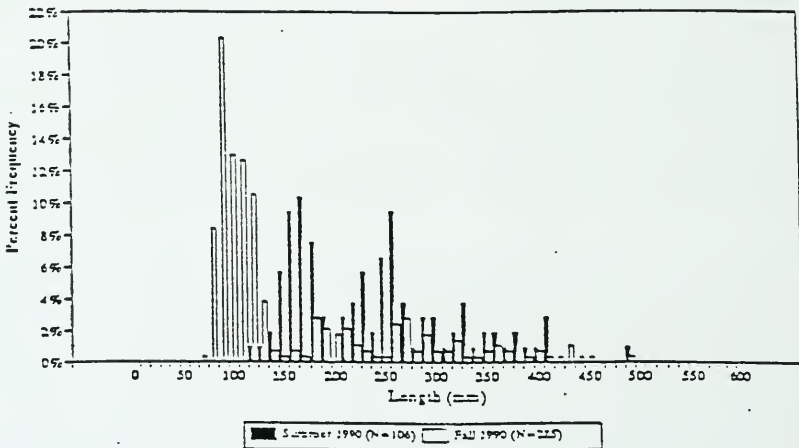


Figure 3b. Length frequency distributions for brown trout collected in summer and fall, from Gold Creek in 1990.

Figure III.1. Figures 3a and 3b from R2 Resource Consultants (1995).

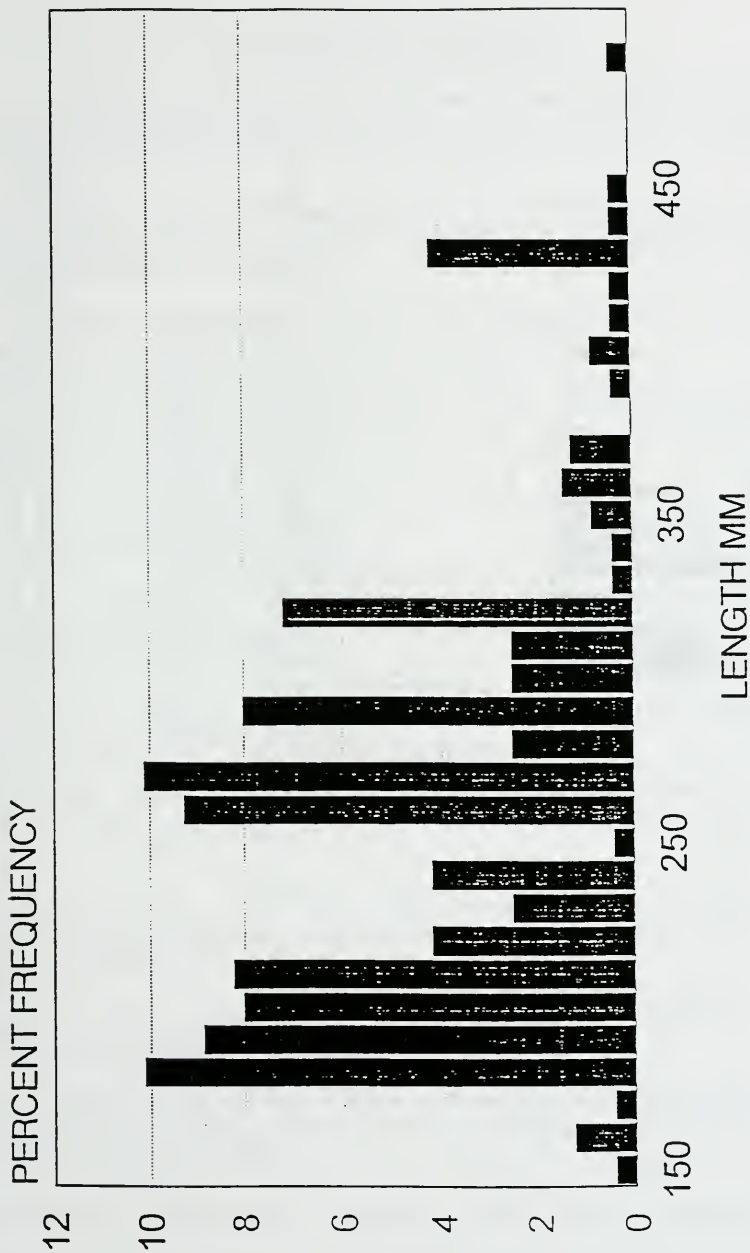


Figure III.2. Length-frequencies of fish 150 mm or larger in fall, 1990, estimated from open bars in R2 Consultants' (1995) Figure 3b (see my Figure 1).



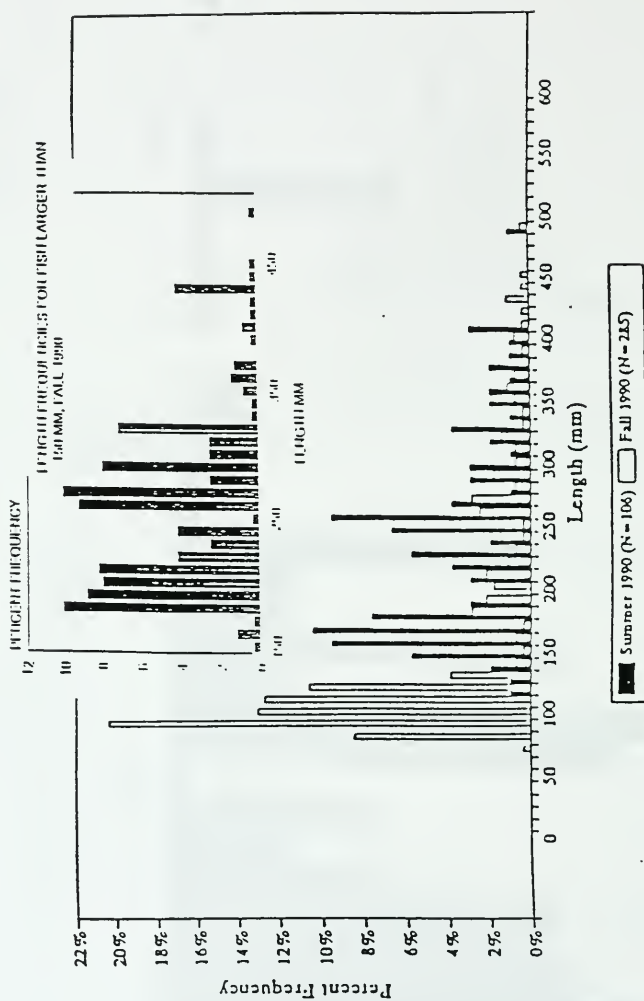


Figure III.3. Figure 2 reduced to scale and superimposed on R2 Consultants' Figure 3b, demonstrating similarity of summer and fall length-frequencies for fish in this size range.

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**APPENDIX E**  
**COMMENTS ON ARCO'S REPORTS CONCERNING THE STATE OF**  
**MONTANA'S INJURY ASSESSMENT FOR THE CLARK FORK RIVER:**  
**EVALUATION OF SNORKEL METHODS**





COMMENTS ON ARCO'S REPORTS  
CONCERNING THE STATE OF MONTANA'S  
INJURY ASSESSMENT FOR THE CLARK FORK RIVER:  
*EVALUATION OF SNORKEL METHODS*

Prepared for

Montana Department of Justice  
Natural Resource Damage Litigation Program  
Old Livestock Building  
1310 E. Lockey Ave.  
Helena, MT 59620

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OCT 04 1995

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DAMAGE PROGRAM

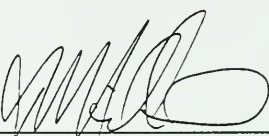
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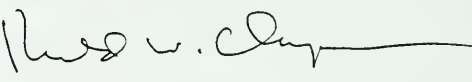
3653 Rickenbacker, Ste 200 • Boise, Idaho 83705  
(208) 383-3401 • FAX (208) 344-4861

September 1995

The following scientists participated in the completion of this work:

A stylized, cursive handwritten signature in black ink, consisting of several loops and a long horizontal stroke at the end.

Tracy W. Hillman

A cursive handwritten signature in black ink, with a long horizontal line extending to the right.

Donald W. Chapman

## Evaluation of Snorkel Methods

In this assessment we compared snorkel counts during daytime and nighttime in the Clark Fork River and Rock Creek. We also compared numbers of trout observed during two-pass (with 3-man crew) and single-pass (with 5-man crew) snorkel surveys in Rock Creek. We conducted surveys between 4 and 8 September 1995.

To compare daytime and nighttime counts, we surveyed one site within each of reaches 1, 2, and 6 on the Clark Fork River and reach 1 on Rock Creek. We selected sites based on ease of access and habitat diversity. We sampled an additional site in reach 6 that contained quiet water and sand/silt substrate. It has been our experience that this quiet water habitat is used frequently by salmonids at night. Thus, we surveyed four sites on the Clark Fork and one on Rock Creek. Each site was 100-m long. Depending on stream width and water clarity, a four or five-man crew surveyed the sites with snorkel gear. We conducted daytime observations between 0900 and 1700 hours. We snorkeled at night between 2100 and 0200 hours. At night we used underwater dive lights to increase visibility. In one site in reach 6 we made two nighttime surveys, one with red-filtered lights and the other without filters. We assumed that red-filtered lights would be less startling to fish and hence may produce a larger population estimate. We surveyed all other sites with unfiltered lights. Daytime and nighttime observations in a given site were within the same 24-hr period. Observers identified trout and assigned them into size classes, each two inches long, from 1 to 25 inches. Water temperatures during daytime surveys ranged from 18.5 to 20°C, while nighttime temperatures ranged from 16 to 17.5°C.

To compare numbers observed during two-pass and single-pass surveys, we selected one pool (45 m long) and one riffle (100 m long) on Rock Creek. We sampled in Rock Creek because numbers of trout were greater there than in the Clark Fork. We selected sites that were wide enough for five observers to make a complete census with one pass. Sites were first surveyed with a 3-man crew.

After a few minutes, we resurveyed the sites with a 5-man crew. Except for the midstream observer in the two-pass method, no other observer snorkeled the same lane twice during the comparison of methods. We surveyed between 1000 and 1700 hours. Observers identified trout and assigned them into size classes. Water temperatures during these surveys ranged from 14.5 to 16°C.

In general, we found that nighttime counts were less than daytime counts in both the Clark Fork River and Rock Creek (Table 1; Appendix A). In only one site (reach 6, state 1/2, site 4) did we count more trout at night than during the day. In that site we also counted fewer trout at night when we used red-filtered lights than when we used unfiltered lights. These results are in contrast with those of ARCO's consultants, who found that nighttime counts were consistently greater than daytime estimates.

Although we do not know the specifics of ARCO's day/night sampling (e.g., snorkeling experience, sampling effort, type of dive lights, etc.), there appear to be at least two reasons why our results differed from those of ARCO's. First, ARCO conducted their surveys in June when water temperatures are colder than in July, August, and early September. At temperatures near 10°C, salmonids, especially small trout (<8 in), conceal themselves during daylight hours, but emerge at night. At these colder temperatures observers may see more trout during the night than during daytime. At warmer water temperatures, when we did our observations, trout are more active and visible during daylight hours. Thus, at warmer temperatures, trout can be seen more easily during the daytime than at night. Second, ARCO indicates that in areas where total coverage was difficult, snorkelers focused their efforts along the stream banks. Therefore, they probably missed trout not stationed near the banks. This would be most apparent in June when streamflows are high and water clarity is typically poor. We were unable to survey most sites in June 1994 because of poor water clarity. Salmonids that use mid-stream locations during the day generally move inshore to shallow, quiescent water at night and rest on or near the substrate. Those fish that used mid-stream locations and were not observed by snorkelers during the day would likely be seen at night in shallow water near the stream edge. Thus, water temperatures or water

clarity or both may explain why ARCO observed more fish at night than during the day.

There are several reasons why nighttime counts during the summer are less reliable than daytime counts. First, some trout tend to move into and out of the light beam. This can result in an overestimate because some fish are counted more than once. For example, during the daytime census we observed a seven-inch brown trout in reach 6 that bore a possible burn mark from exposure to electrofishing gear. That fish was observed four different times during the night by two different snorkelers. It is likely that fish without unique marks were repeatedly counted during nighttime observations. Second, some fish are instantly startled by the light beam. As some trout do, whitefish, squawfish, and suckers often bolt in all directions making it very difficult to identify them. Both whitefish and squawfish look very much like trout when they bolt from the light beam. Thus, under the same conditions an observer is more likely to misidentify a fish at night than during the day. Furthermore, when fish move off rapidly in all directions, there is little accountability of fish moving across different counting lanes. Third, nighttime observations are more dangerous than daytime observations. Because the field of view is restricted at night, observers frequently encounter snags. This occurs most often along the stream edge where sticks, brush, stumps, large angular rock, rebar, car-body parts, cable, and other debris stab, puncture, or entangle observers. We have observers with several hundred hours of nighttime snorkeling experience that have also encountered angry beaver, otter, and rattlesnakes in the water at night. Such encounters tend to diminish the observer's ability to count fish. Finally, we have found it very difficult to acquire permission from landowners to access their property late at night.

We found little difference between numbers of trout observed during two-pass (3-man crew) and single-pass (5-man crew) surveys (Table 2; Appendix B). In the riffle site, the single-pass method estimated two more fish than the two-pass method. In the pool, however, the two-pass method estimated two more fish than the single-pass method. These results suggest that there is no consistent bias in the two methods.

Table 1. Comparison of numbers of trout observed during day and night snorkel surveys in the Clark Fork River and Rock Creek during September 1995.

Stream	Reach	State	Site	Time of Survey	Size classes (inches)														Total
					1	3	5	7	9	11	13	15	17	19					
Clark Fk	1	3/5	4	Day			8	10	1	1	2					22			
				Night		1	5	1			1					8			
Clark Fk	2	3	4	Day			1		2	1						4			
				Night					1							1			
Clark Fk	6	1/2	4	Day		1	1	5		1	1	2	1			12			
				Night <sup>1</sup>			5				1		1			7			
				Night		4	5	5	3	1	2					20			
Clark Fk	6	1/2	4 <sup>2</sup>	Day		2	8	4		2	1					17			
				Night			7	4	2		1					14			
Rock Ck	1	3	2	Day		21	38	11	5	1	1	1	3	1		80			
				Night		11	7	4	2	2	2	5	4	1		38			

<sup>1</sup>Nighttime counts conducted with red-filtered lights.

<sup>2</sup>Site located about 40 m downstream from site #4.

Stream	Reach	State	Site	Type of Survey	Size classes (inches)													Total
					1	3	5	7	9	11	13	15	17	19				
Rock Ck	1	3	3 (riffle)	2-pass			3	4		2	1					10		
				1-pass			4	4		2	1	1				12		
Rock Ck	1	3	7 (pool)	2-pass			8	5	4	2						19		
				1-pass			7	5	3	2						17		

Appendix A. Field data sheets of numbers of trout observed during day and night snorkeling in the Clark Fork River and Rock Creek, September 1995.



Date 9/5/95  
Temp. 19 : 12.5  
Obs. IF KH, DM, MM, TH

1: Daytime

2. = Nichtline

Size in inches (total lengths)

[illegible][illegible][illegible]

Reach Fevertail

Date

9/5/95

State  $R2: S3 = 4$

Temp.

 $20:12$ 

Areas 0.32

Obs.

IR. 0m, mm, KH, TH

L = Daytime

2 = Nighttime

Size in inches (total lengths)

[illegible][illegible][illegible][illegible]

Date 9/6/95

Temp. 20 : 17

Obs. NM, MM IR MM

1 = Daytime      2 = Red Filtered Night      3 = Unfiltered Night

Size in inches (total lengths)

[illegible][illegible][illegible][illegible]

Reach Galen = 2

Date 2/6/95

State F.6:  $\leq 1/2$  below = 4

Temp. 20; 17

Areas 0.13

Obs. Um, mm, IR mm

1 = Daytime      2 = Nighttime

Size in inches (total lengths)

[illegible][illegible][illegible][illegible]

$1 = \text{Day Line}$        $2 = \text{Night Line}$

---

Appendix B. Field data sheets of numbers of trout observed during two-pass (3-man crew) and single-pass (5-man crew) snorkel surveys in Rock Creek, September 1995.

Reach Rock Creek (R-221e) Date 9/7/95  
 State SE  $\bar{\Delta}$  = 1 Temp. 14.5°C  
 Areas 0.19 Obs. JP, NW, KH, MM, TH

1 = 3-man crew (2 pass) 2 = 5-man crew  
Size in inches (total lengths)

[illegible][illegible][illegible][illegible]



Reach Rock Creek (Pool) Date 9/7/95  
 State SE: =2 Temp. 16°C  
 Areas 209 Obs. IR, UM, KH, MM, TH  
 1 = 3-man crew (2-pass) 2 = 5-man crew (1-pass)

Size in inches (total lengths)

		1	3	5	7	9	11	13	15	17	19	21	23	25	27	Total
Brown	1			5	3	2	1									11
	2			6	3	2	1									12
	3															
	4															
Total																

Rainbow	1			3	2	2	1									8
	2			1	2	1	1									5
	3															
	4															
Total																

Bull	1															
	2															
	3															
	4															
Total																

	1															
	2															
	3															
	4															
Total																

19/17







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**APPENDIX F**  
**RESPONSE TO ARCO'S REPORTS CONCERNING THE STATE OF**  
**MONTANA'S INJURY ASSESSMENT OF THE UPPER CLARK FORK**  
**RIVER BASIN**



**RESPONSE TO ARCO'S REPORTS  
CONCERNING THE STATE OF MONTANA'S  
INJURY ASSESSMENT  
OF THE  
UPPER CLARK FORK RIVER BASIN**

*Prepared for:*

**State of Montana**

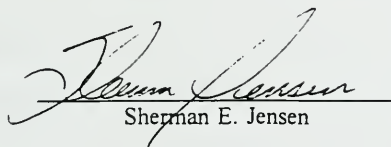
*Prepared by:*

**Sherman Jensen  
White Horse Associates  
Logan, Utah**

**September, 1995**



The following scientists participated in the completion of this work:



Handwritten signature of Sherman E. Jensen, written in cursive script, positioned above a horizontal line.

Sherman E. Jensen





## 1.0 INTRODUCTION

This document addresses major themes of expert reports prepared by Dudley Reiser, Ph.D. and Thomas Wesche, Ph.D for the Atlantic Richfield Company (ARCO) concerning the Natural Resource Damage Assessment conducted by the State of Montana for the upper Clark Fork River (CFR)<sup>1</sup>.

## 2.0 TASKS

The tasks addressed are:

1. Quantify the extent of stream alteration for the CFR and Silver Bow Creek (SBC).
2. Quantify the extent of land uses in the CFR and reference basins.
3. Evaluate the extent of confinement for straight reaches of reference streams.
4. Evaluate Reiser's criticism of the use of Ecoregion as the broadest level of the hierarchical classification.

Results are presented with respect to each task.

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<sup>1</sup> In the course of this work, two errors in the Don Chapman report dated January, 1995 were found:

1. The river miles stated in Table 3 (page 38) for Reach 6 of the CFR are not correct. The first six states, as listed in the table are correct. The seventh listed state (Eroded banks [2]) should be river miles 97.76-106.75. The river miles for all subsequent states of Reach 6, 7, 8, 9 and 10 should be that listed, plus (+) 6.08 miles. This same error on the Clark Fork River - Maps 35 through 49, which were not submitted as part of the report. I do not believe that this has affected any of the calculations of fish density because they were determined from a measure of stream area determined from the GIS.

2. The Landtype Associations listed in Appendix Table B5 (page 75) should be "Fluvial Lands" for both Reach 5 of the Clark Fork and Reach 4 Big Hole.

## 2.1 Stream Alteration

In both the Wesche and Reiser reports the extent of channelization in the Clark Fork River relative to that in reference streams is a major point of contention. On page 3 (paragraph 3)

Wesche states:

"Channel alterations, including realignment, straightening, meander cutoffs, confinement and rip-rapping... are common on the CFR from Warm Springs Ponds to Milltown Reservoir. I estimate that approximately 60 miles (48%) of the CFR in this section have been altered."

On page C-26 Reiser states:

"The effects of channelization have included the actual loss of about 18 miles of the lower river resulting from the cutting off of channel meanders as part of the straightening process. Collectively about 50 miles of the CFR have been channelized, or about 50 percent of its length between Missoula and Warm Springs."

The objective of this task was to refine estimates of the lengths of the CFR and SBC that have been altered through realignment, straightening, meander cutoffs, confinement and rip-rapping. The approach entailed:

1. Streambanks that have been altered by transportation and/or cultural development were identified from 1:8,000 scale color infrared (CIR) aerial photos dated August, 1983. Altered streambanks were further classified into three classes:

- A. Confined Unvegetated: Stream course is confined and shrub/tree vegetation is sparse or absent on streambank.
- B. Confined Vegetated: Stream course is confined and shrub/tree vegetation is present on streambank.
- C. Unconfined Rip-rap: Stream course is not confined by transportation and/or cultural development but streambanks have been rip-rapped.

Classes of altered streambanks were marked on 1:24,000 scale USGS quads. For the CFR, which is a polygon on the quads, altered streambanks were marked on the right and left bank (facing downstream) as appropriate. For SBC, which is a line on the quads, an attribute was used to identify reaches where the left, right or both streambanks were altered. The CFR, SBC and altered streambanks were digitized from the 1:24,000 scale

quads. The lengths of not-confined, confined unvegetated, confined vegetated and unconfined rip-rapped were summarized by stream reach and for all reaches of the CFR and SBC.

2. Realigned reaches have been moved to accommodate transportation routes or in response to natural processes. Reaches of the CFR that may have been realigned to accommodate transportation and/or cultural development were identified from 1:8,000 CIR aerial photos dated August, 1983. Old river channels were generally apparent on the aerial photos. Some of these may have been overflow channels of the CFR channel and not realigned to accommodate cultural features. The center of stream courses prior to realignment were marked on 1:24,000 scale USGS quads. The center-line of the CFR before realignment and after realignment were digitized from these quads. Results are summarized by reach for the CFR. This approach could not be used for SBC because the old stream channels were generally obliterated by tailings.

3. The total length of the CFR that has been altered was estimated by reach as the length of realigned stream channel plus the length of altered streambank that was not realigned.

Results of the analysis of streambank alteration are summarized by reach in Table 2.1-1 and for all reaches of the CFR and SBC in Table 2.1-2. Results of the analysis of channel realignment of the CFR are summarized by reach in Table 2.1-3. The total length of the CFR that has been altered is summarized by reach in Table 2.1-4.

Results indicate that about 33.3 miles (12.5 percent) of the streambanks along the CFR have been altered through confinement or rip-rap. Of this 33.3 miles, 18.3 miles was also realigned and 15.0 miles was not realigned. About 1.4 miles that was not realigned was *sinuous* streambank in agricultural fields that had been rip-rapped. About 25.3 miles of the CFR has been realigned. The total length of the river has been reduced by about 6.1 miles (4.8 percent of the length before realignment). Estimated as the sum of the length of the CFR that is realigned plus the length of streambank that is confined, but not realigned, a total of 40.2 miles (33.2 percent) of the CFR have been altered. The estimates of stream alteration presented by Wesche (60 miles or 48 percent) and Reiser (50 miles or 50 percent) are greater than were found in this study.

About 16.6 miles (27 percent) of the streambanks along SBC are confined. Much of the SBC channel is confined by mine tailings. Most of Reach 7 of SBC has been realigned. An exception is the "eroded bank" state identified in the lower part of Reach 7, which has a sinuosity of 1.72. Extrapolating this sinuosity for all of Reach 7, it has been shortened by about 6.0 miles in response to realignment. Loss of stream length was not considered for Reach 8 and 9 of SBC. The sinuosity of Reach 10 SBC is expected to have been similar to Reach 3 of Bison Creek (1.49). Extrapolating this sinuosity to Reach 10 of SBC, it has been shortened by about 3.8 miles. The total lengths of Reach 7 and 10 of SBC have been reduced by 9.8 miles or 29.3 percent of the length before realignment. Most of SBC is altered. The existing conditions are conducive to erosion and transport of tailings.

Table 2.1-1. Altered streambank summary by reach.

STREAM	REACH	ALTERED CLASS	-----LENGTH----- (miles)	(%)
CLARKFK	1	NOT CONFINED	40.6	90.1
CLARKFK	1	CONFINED-UNVEGETATED	3.9	8.7
CLARKFK	1	CONFINED-VEGETATED	0.6	1.3
CLARKFK	1	TOTAL	45.1	100.0
CLARKFK	2	NOT CONFINED	45.7	72.5
CLARKFK	2	CONFINED-UNVEGETATED	12.1	19.2
CLARKFK	2	CONFINED-VEGETATED	3.9	6.1
CLARKFK	2	RIP-RAP BUT NOT CONFINED	1.4	2.2
CLARKFK	2	TOTAL	63.0	100.0
CLARKFK	3	NOT CONFINED	8.1	89.4
CLARKFK	3	CONFINED-UNVEGETATED	0.7	7.7
CLARKFK	3	CONFINED-VEGETATED	0.3	2.9
CLARKFK	3	TOTAL	9.1	100.0
CLARKFK	4	NOT CONFINED	41.7	95.3
CLARKFK	4	CONFINED-UNVEGETATED	2.1	4.7
CLARKFK	4	TOTAL	43.8	100.0
CLARKFK	5	NOT CONFINED	12.7	71.1
CLARKFK	5	CONFINED-UNVEGETATED	3.7	20.6
CLARKFK	5	CONFINED-VEGETATED	1.5	8.3
CLARKFK	5	TOTAL	17.8	100.0
CLARKFK	6	NOT CONFINED	83.9	96.3
CLARKFK	6	CONFINED-UNVEGETATED	1.1	1.3
CLARKFK	6	CONFINED-TAILINGS.	2.1	2.4
CLARKFK	6	TOTAL	87.2	100.0
SILVER BOW	7	NOT CONFINED	21.9	82.6
SILVER BOW	7	BOTH BANKS CONFINED	4.3	16.1
SILVER BOW	7	RIGHT BANK CONFINED	0.1	0.5
SILVER BOW	7	LEFT BANK CONFINED	0.2	0.9
SILVER BOW	7	TOTAL	26.5	100.0
SILVER BOW	8&9	NOT CONFINED	3.7	47.5
SILVER BOW	8&9	BOTH BANKS CONFINED	3.5	44.7
SILVER BOW	8&9	RIGHT BANK CONFINED	0.4	5.7
SILVER BOW	8&9	LEFT BANK CONFINED	0.2	2.0
SILVER BOW	8&9	TOTAL	7.7	100.0
SILVER BOW	10	NOT CONFINED	19.2	70.8
SILVER BOW	10	BOTH BANKS CONFINED	7.1	26.3
SILVER BOW	10	RIGHT BANK CONFINED	0.7	2.5
SILVER BOW	10	LEFT BANK CONFINED	0.1	0.4
SILVER BOW	10	TOTAL	27.1	100.0

Table 2.1-2. Altered streambank summary for CFR and SBC.

STREAM	ALTERED CLASS	-----LENGTH-----	
		(miles)	(%)
CLARKFK	NOT CONFINED	232.7	87.5
CLARKFK	CONFINED-UNVEGETATED	23.6	8.9
CLARKFK	CONFINED-VEGETATED	6.2	2.3
CLARKFK	CONFINED-TAILINGS.	2.1	0.8
CLARKFK	RIP-RAP BUT NOT CONFINED	1.4	0.5
	TOTAL	266.0	100.0
SILVER BOW	NOT CONFINED	44.7	72.9
SILVER BOW	BOTH BANKS CONFINED	14.9	24.2
SILVER BOW	RIGHT BANK CONFINED	1.2	2.0
SILVER BOW	LEFT BANK CONFINED	0.5	0.8
	TOTAL	61.3	100.0

Table 2.1-3. Realignment summary by reach (units are miles unless otherwise specified).

REACH	Not Realigned (1)	Realigned After (2)	Total After Realignment (3)	Realigned Before (4)	Total Before Realignment (5)	Total Difference (miles) (6)	Total Difference (%) (7)
1	11.9	4.4	16.4	3.6	15.6	0.8	5.1
2	17.7	13.3	30.9	17.8	35.4	-4.5	-12.7
3	2.7	1.4	4.1	2.4	5.1	-1.0	-19.1
4	18.0	1.8	19.8	2.1	20.1	-0.3	-1.7
5	4.2	4.3	8.6	5.4	9.6	-1.1	-11.0
6	41.2	0.1	41.3	0.1	41.4	-0.1	-0.2
<b>TOTAL</b>	<b>95.8</b>	<b>2.0</b>	<b>121.1</b>	<b>31.4</b>	<b>127.2</b>	<b>-6.1</b>	<b>-4.8</b>

(1) Length of stream (miles) that was not realigned.

(2) Length of stream (miles) that was realigned (after realignment).

(3) Total length of stream (miles) after realignment (1 + 2)

(4) Length of stream (miles) that was realigned (before realignment).

(5) Total length of stream (miles) before realignment (1 + 4).

(6) Difference in total length (miles) of stream before and after realignment (3 - 5).

(7) Percent difference in stream length (6/5 \* 100%).

Table 2.1-4. Altered stream summary by reach.

REACH	TOTAL LENGTH (1)	-----CONFINEMENT CLASS----- Unvegetated (2)	Vegetated (3)	Rip-Rap (4)	REALIGNED (5)	TOTAL ALTERED (6)	PERCENT ALTERED (7)
1	16.4	1.8	0.6	0.0	4.4	6.8	41.6
2	30.9	4.1	1.2	1.0	13.3	19.6	63.3
3	4.1	0.3	0.3	0.0	1.4	1.9	47.4
4	19.8	1.1	0.0	0.0	1.8	2.9	14.6
5	8.6	1.9	0.4	0.0	4.3	6.6	77.5
6	41.3	1.1	1.2	0.0	0.1	2.3	5.7
TOTAL	121.1	10.4	3.6	1.0	25.2	40.2	33.2

- (1) Total length of stream reach (miles).
- (2) Length of unvegetated confined streambank (miles) that is not realigned.
- (3) Length of vegetated confined streambank (miles) that is not realigned.
- (4) Length of sinuous, rip-raped streambank (miles) that is not realigned.
- (5) Length of realigned channel (miles).
- (6) Total length (miles) of altered channel (2 + 3 + 4 + 5).
- (7) Percent of total length (1) that is altered ( $5/1 * 100\%$ ).



## 2.2 Land Uses

Both Wesche and Reiser refer to differences in land-use between the CFR basin and reference basins. Wesche considered amount of irrigated lands and channel alterations. He estimated acres of irrigated land from the USGS Surface Water Records for Montana (1993).

Reiser (page D2-60) discusses land use and vegetation cover digital data from the State of Montana's Natural Resource Information System (NRIS). He states:

"Geographical Information System (GIS) coverages of land use and vegetation were provided at a scale of 1:250,000. The Beaverhead, Big Hole, and Ruby Rivers were provided by NRIS on a county basis at a scale of 1:100,000. The Clark Fork River, Silver Bow Creek, Flint Creek and Rock Creek were digitized from quadrangles at a scale of 1:24,000.

On page D2-65 Reiser says:

"Physical, chemical, and biologic influences on trout population were evaluated at three general scales: at the watershed/drainage basin scale, at the reach level, and at the local site level. The type and nature of each influence varies with the scale at which a particular factor acts, be it at the drainage basin, reach, or local site level."

Reiser presents combined land use and vegetation types within the riparian corridors of the CFR and references reaches in Table 2.2-11. Reiser does not state how the "riparian corridor" was defined. Given the scale of the GIS coverages (1:250,000 scale), it may be inappropriate to use them to identify combined land use and vegetation types within a narrow stream/riparian corridor. It is more appropriate to evaluate similarities in land use at the Ecoregion/drainage basin scale.

Ecoregion is the broadest level of the hierarchical classification used by the State of Montana to stratify basins into homogeneous areas. Land use and potential natural vegetation were used, in part, to identify Ecoregions (Omernik 1987). Thus it is expected that land use and vegetation in the Montana Valley and Foothill Prairie (MVFP) Ecoregion in the CFR basin will be similar to land use and vegetation in the MVFP Ecoregion in reference basins. Analyses of

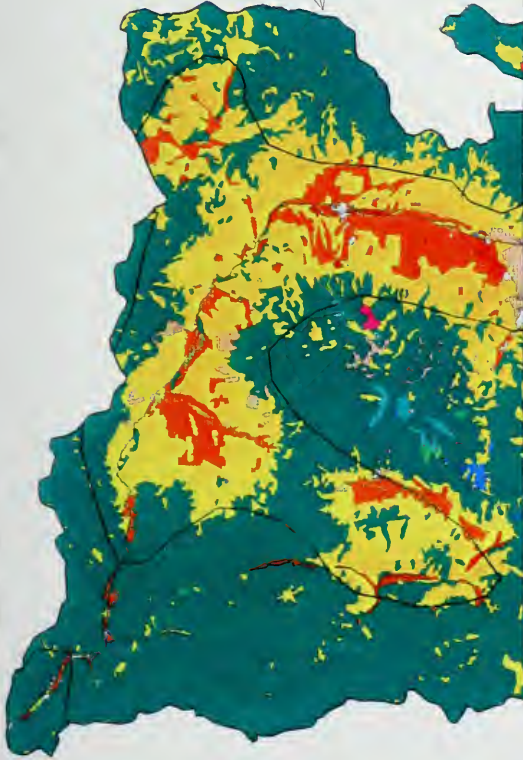
the distribution of land use for Ecoregions in the CFR and reference basins were conducted.

The 1:250,000 scale GIS map coverage of land use and vegetation was acquired from USGS. Land use was evaluated for: 1) Ecoregions in southwest Montana; and 2) Ecoregions of CFR and reference basins. The CFR basin was digitized from 1:24,000 scale topographic maps. Other watersheds were digitized from 1:250,000 scale topographic maps.

A map showing land use relative to Ecoregions (Omernik 1987) is presented as Figure 2.2-1. The distribution of land use by Ecoregion is depicted in Figure 2.2-2. Dominant land uses in the Northern and Middle Rockies Ecoregions are range and forest. Range is the dominant land use in the Montana Valley and Foothill Prairies Ecoregion, complimented by sizeable proportions of forest and agriculture.

Montana  
Valley  
and  
Foothill  
Praries

Northern  
Rockies



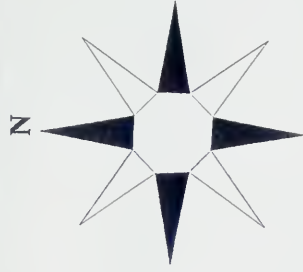
## Ecoregions/Landuses Control and Reference Basins

Figure 2.2-1

*Note: Landuse from 1:250,000 Scale USGS  
Digital Landuse Maps; Ecoregions  
from Omernick (1987)*

### Legend

- Urban
- Agriculture Lands
- Range Lands
- Deciduous Forest Lands
- Evergreen Forest Lands
- Mixed Forest Lands
- Water
- Wetlands
- Barren Lands
- Tundra



Montana  
Valley  
and  
Foothill  
Praries

Northern  
Rockies

Northern  
Rockies

Middle  
Rockies

Montana Valley and  
Foothill Praries

Middle  
Rockies

## Ecoregions/Landuses Control and Reference Basins

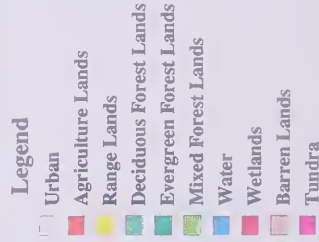


Figure 2.2-1

*Note: Landuses from 1:250,000 Scale USGS  
Digital Land Use Data, Ecoregions  
from Omernick (1987)*

# LAND USE BY ECOREGION

## CFR AND REFERENCE BASINS

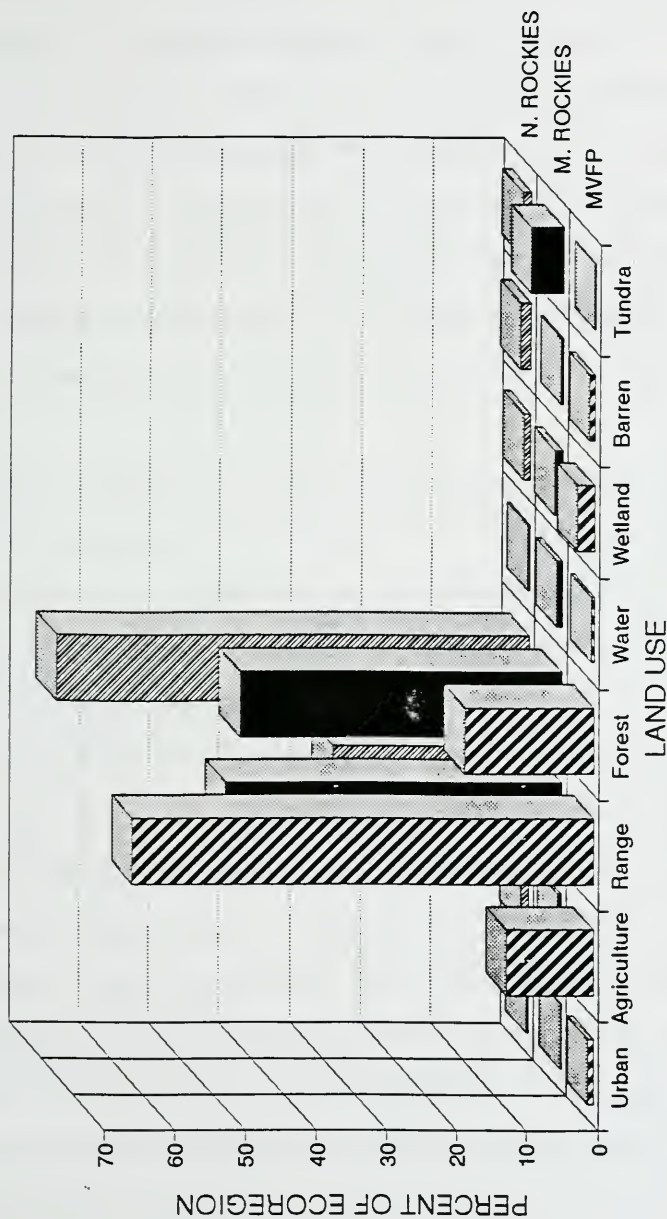


Figure 2.2-2. Land use by Ecoregion, CFR and reference basins.





# LAND USE BY ECOREGION

## CFR AND REFERENCE BASINS

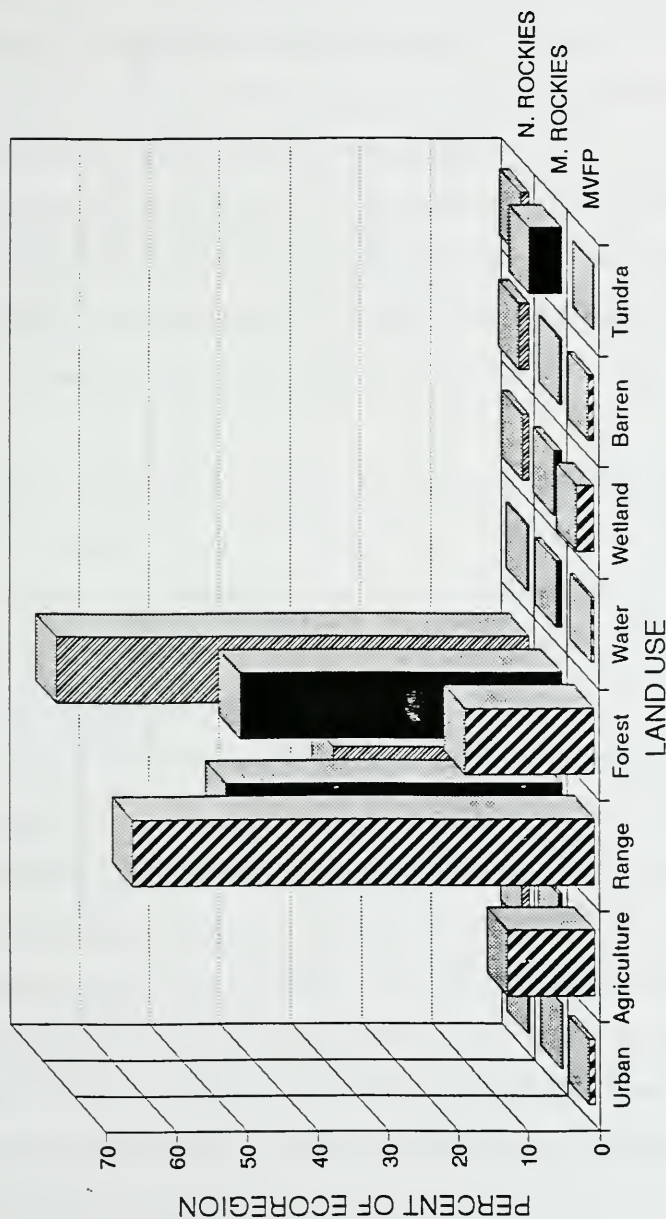


Figure 2.2-2. Land use by Ecoregion, CFR and reference basins.





Land uses in Ecoregions of the CFR and reference basins were also compared. Under the hierarchical classification system, paired CFR and reference sites generally fall within the same Ecoregion. Consequently, it was expected that land uses, which are relevant to the biological potential of aquatic systems, would be similar (see section 2.2 ecoregion). Land uses of the CFR basin in the Northern Rocky Mountain Ecoregion were compared to land uses of Rock Creek basin in the same Ecoregion. Land uses of the CFR basin in the MVFP Ecoregion were compared with land uses of Flint, Big Hole River (BHR), Beaverhead and Ruby Rivers basins in the same Ecoregion. The CFR basin used for this comparison includes Rock, Flint and SBC basins. The areas of land uses in the CFR and reference basins are tabulated by Ecoregion in APPENDIX A.

The distribution of land uses in the CFR and reference basins for the Northern and Middle Rocky Mountains Ecoregions are compared in Figure 2.2-3. The CFR basin includes the watersheds of Rock Creek, Flint Creek and SBC. Land uses in the Northern Rocky Mountains Ecoregion are similar for the CFR and Rock Creek basins. The greatest differences are for range (6 percent higher for CFR) and forest (5.6 percent higher for Rock).

The distribution of land uses in the CFR and reference basins for the MVFP Ecoregion are compared in Figure 2.2-4. Land uses in the MVFP Ecoregion of the CFR basin are most similar to Flint and Big Hole River (BHR) basins. The Beaverhead basin has a higher proportion of range and a corresponding lower proportion of forest than the CFR basin. Other land uses in the Beaverhead basin are similar to the CFR basin. The Ruby basin has a lower proportion of forest and a higher proportion of agricultural land. Other land uses in the Ruby basin are similar to the CFR basin. It is expected that higher proportions of range in the Beaverhead basin and higher proportions of agricultural land in Ruby basin would result in greater impacts to stream resources compared with the CFR basin.

# LAND USE FOR N. ROCKIES ECOREGION

## CLARK FORK VERSUS ROCK BASINS

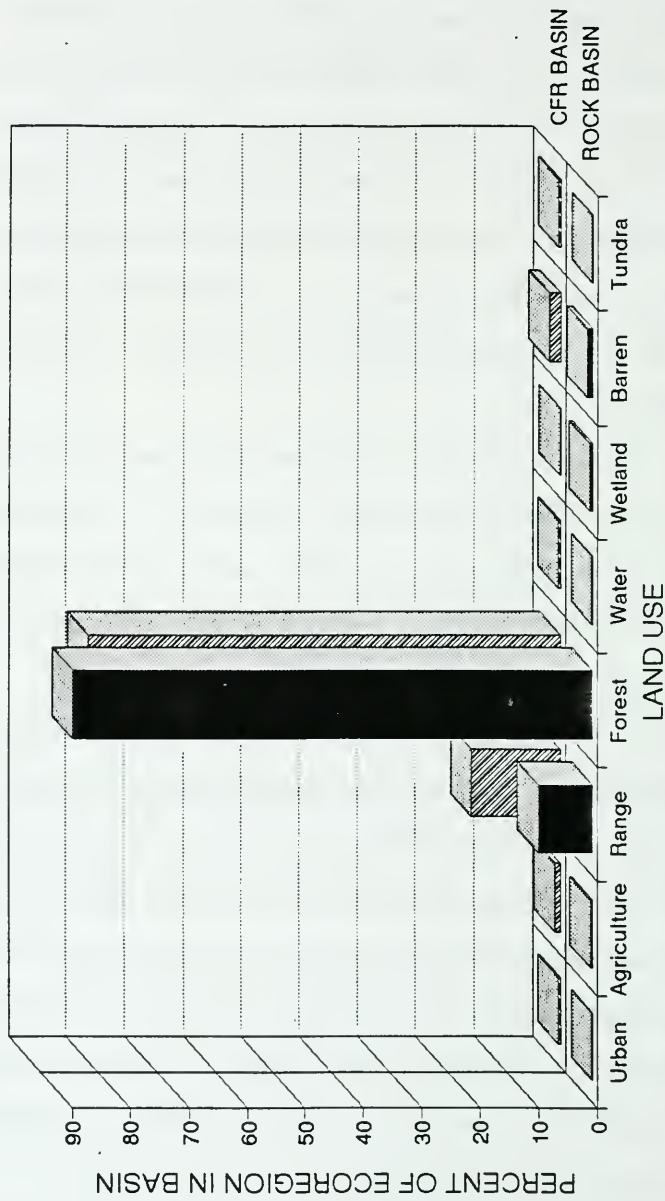


Figure 2.2-3. Land use in the Northern Rocky Mountain Ecoregion, CFR versus Rock Creek basins.

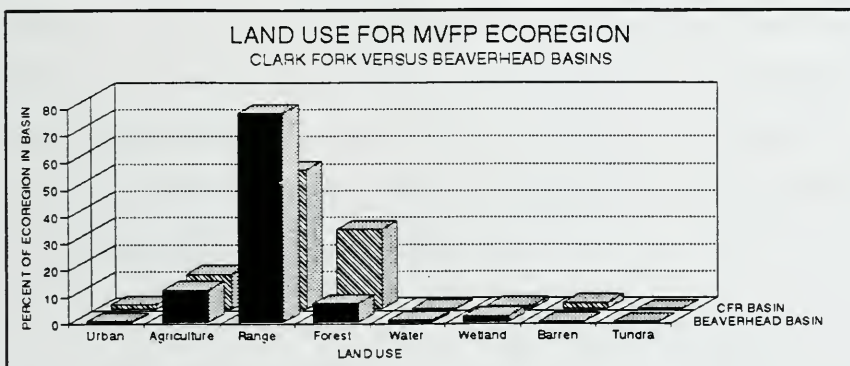
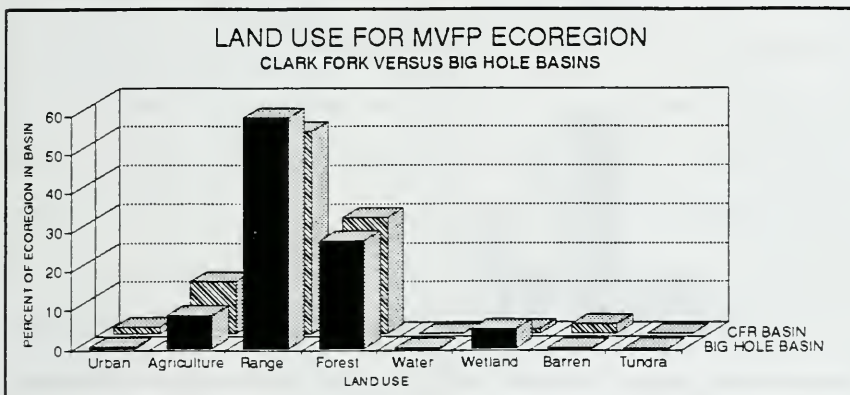
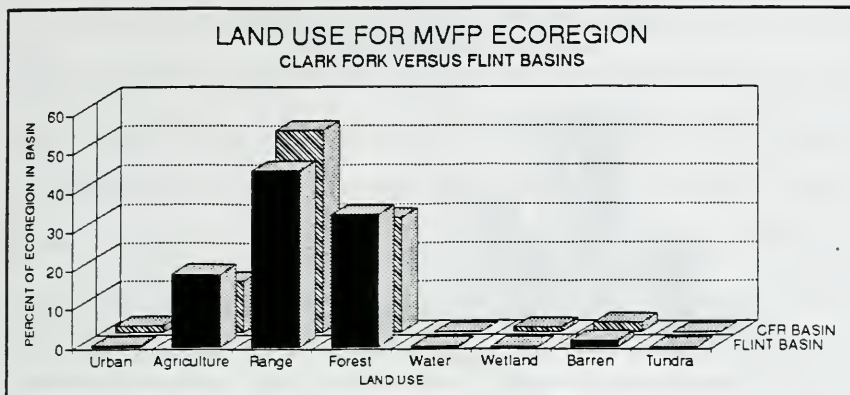


Figure 2.2-4. Land use for Montana Valley and Foothill Prairies Ecoregion, CFR versus reference basins.

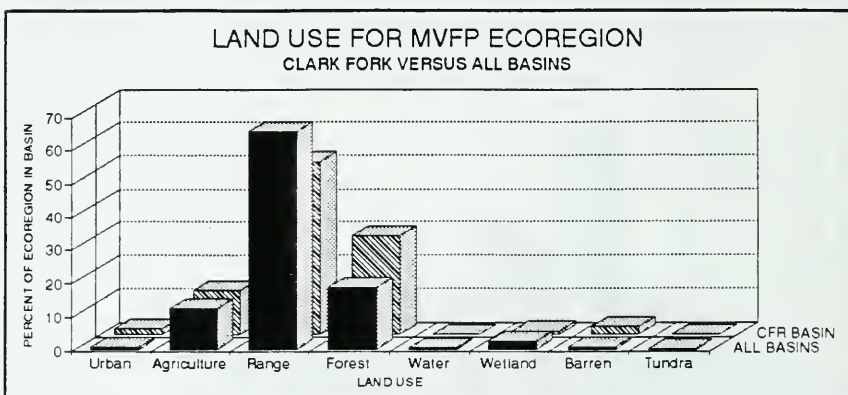
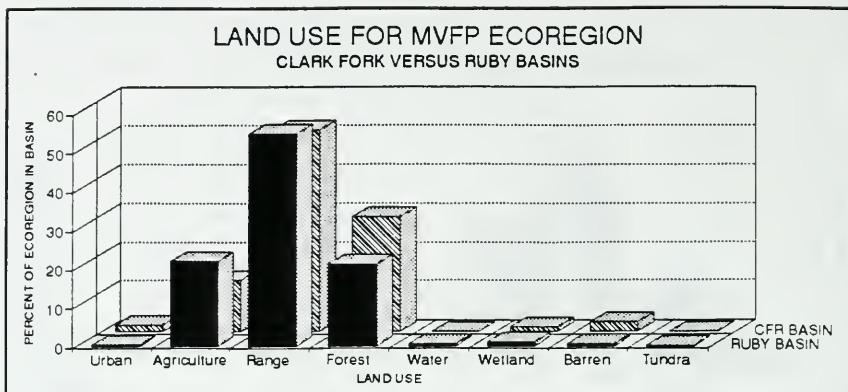


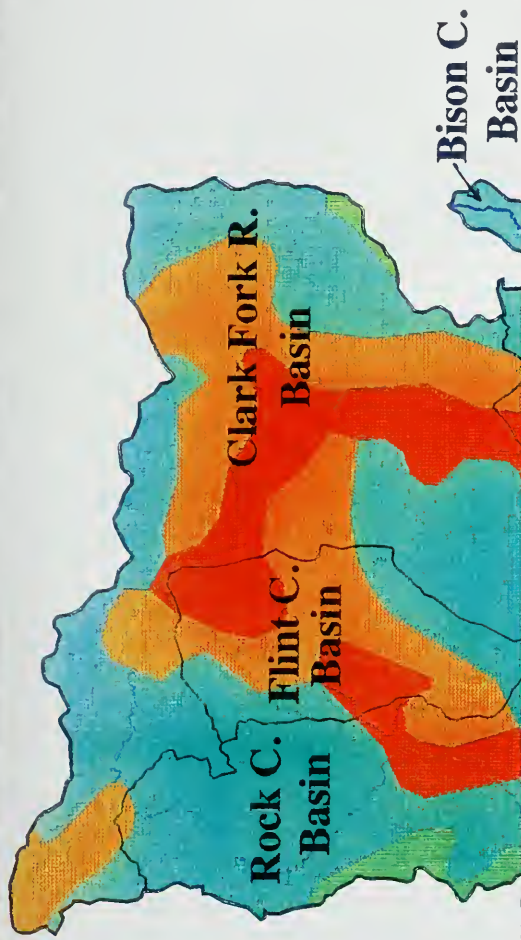
Figure 2.2-4. Continued.

Wesche compares the area (square miles) of irrigated land above stream gages on the Clark Fork River and selected reference streams (Table 43 on page B-72). A more appropriate comparison would be the proportion of the watershed that is irrigated. A digital map file of land use for the state of Montana was obtained from the Natural Resource Information System at the Montana State Library. This land use map was prepared from the USGS National Atlas (1970) and includes "irrigated lands". The watershed boundaries of the Clark Fork River, Rock Creek, Flint Creek and the BHR were overlaid on the digital land use map. A map illustrating land uses from the USGS National Atlas (1970) is presented as Figure 2.2-5.

The areas (square miles) of irrigated land as presented by Wesche versus that estimated by White Horse Associates (WHA) from the USGS National Atlas are compared in the upper graph of Figure 2.2-6. The area of irrigated land reported by Wesche is less than that from the USGS National Atlas. A more meaningful comparison of the proportion of the CFR and reference basins that are irrigated land is shown in the lower graph of Figure 2.2-6. The USGS National Atlas indicates that irrigated lands comprise between 10 and 25 percent of the CFR, Flint Creek, Rock Creek and BHR basins. Flint Creek and the BHR basins have the largest proportions of irrigated lands. Rock Creek basin has about 5 percent less irrigated land than the CFR basin. The proportion of irrigated land from the USGS National Atlas for Flint Creek, Rock Creek and the BHR basins are all within 10 percent of that of the Clark Fork River. Using Wesche's estimates of the area (square miles) of irrigated land, the percent irrigated lands are nearly identical for the CFR, Flint and BHR basins.





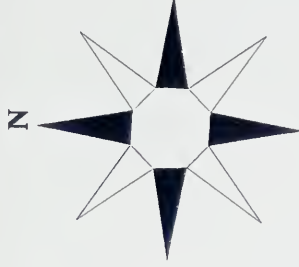


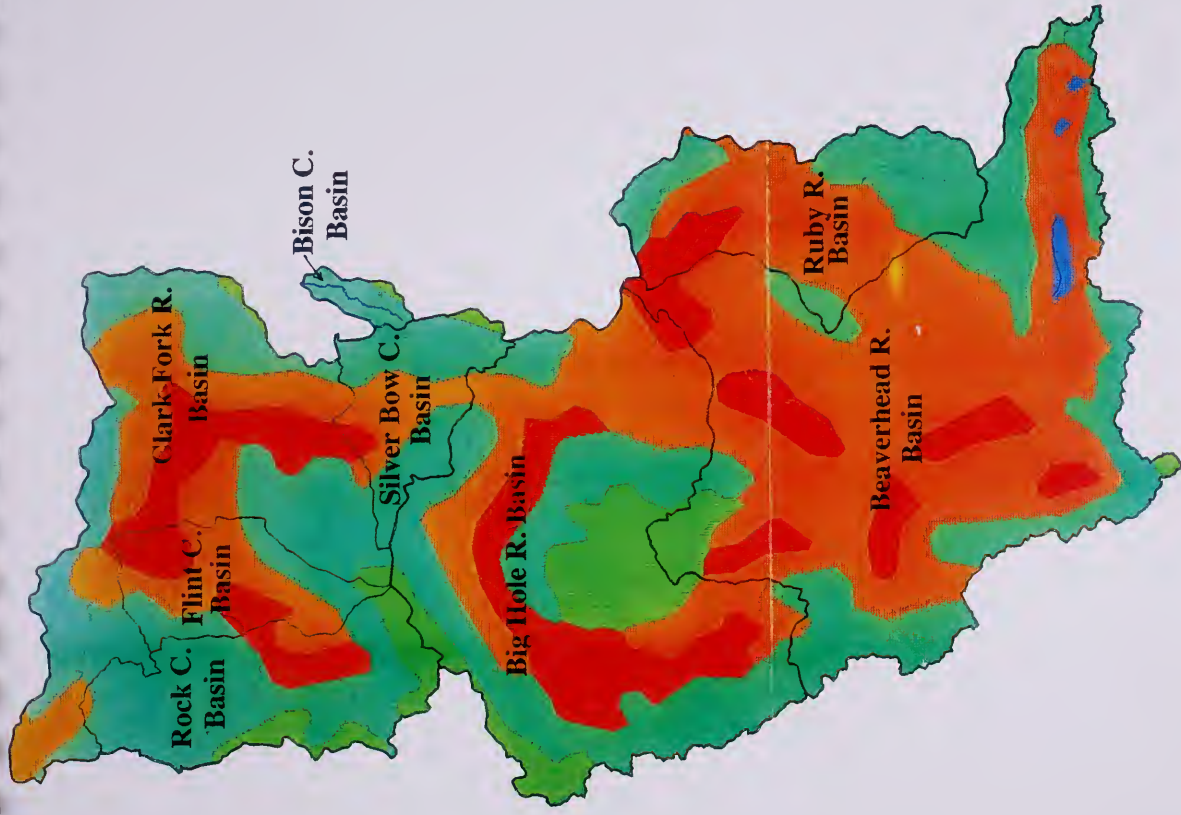
# **Landuses Control and Reference Basins**

**Figure 2.2-5**

## **Legend**

-  Irrigated lands
-  Forest and woodland mostly grazed
-  Forest and woodland mostly ungrazed
-  Subhumid grassland and semiarid grazing land
-  Desert shrubland grazed
-  Open water





## Landuses Control and Reference Basins

### Legend

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- Desert shrubland grazed
- Open water



0 30  
Miles

Figure 2.2-5



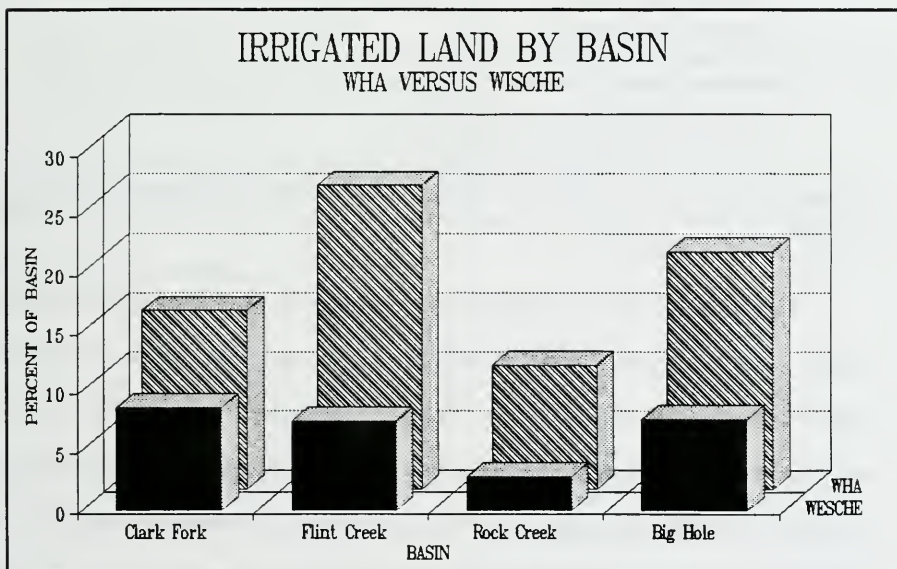
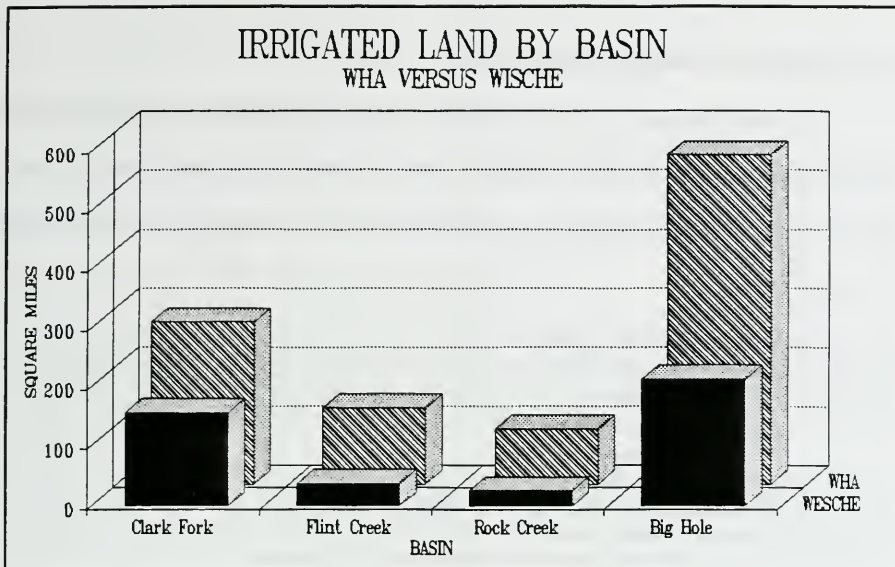


Figure 2.2-6. Comparison of areas of irrigated land estimated by White Horse Associates (WHA) and Wesche.



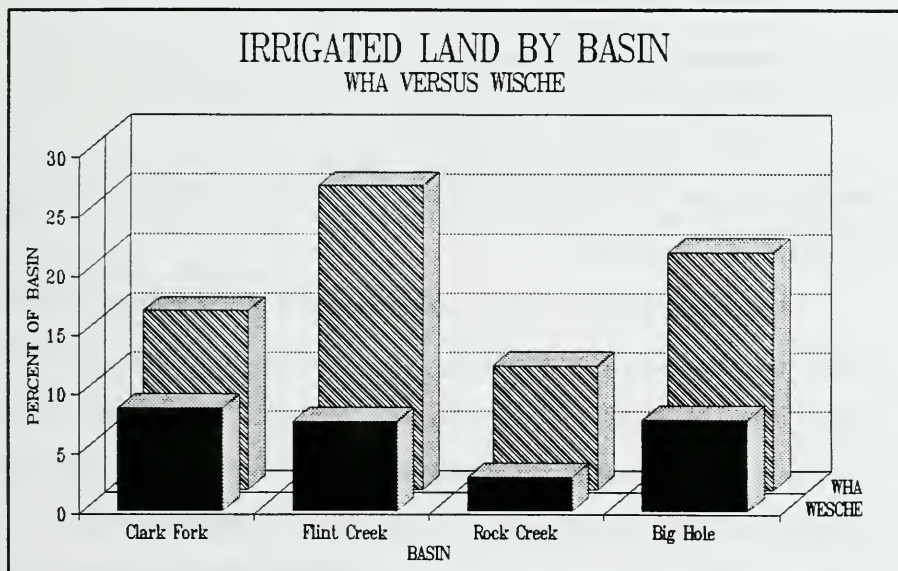
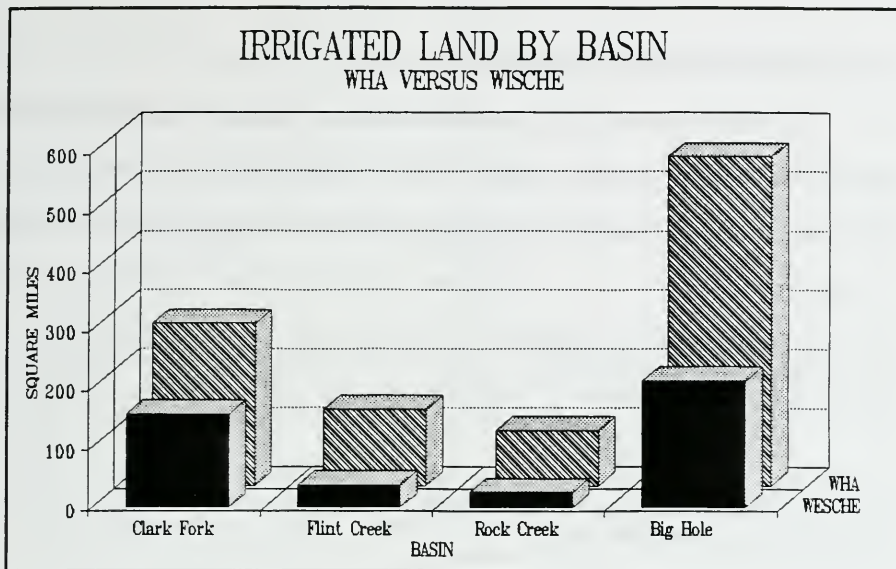


Figure 2.2-6. Comparison of areas of irrigated land estimated by White Horse Associates (WHA) and Wesche.

## 2.3 Straight Reference Reaches

Wesche discusses the effects of stream alteration and contrast channelized reaches of the CFR with "straight" reaches of reference streams. Wesche (page B-3) lists hydraulic and morphologic changes resulting from confining a stream to a straight course in response to channelization:

- \* reduced channel length;
- \* increased channel slope;
- \* reduced sinuosity;
- \* increased sediment transport;
- \* streambed degradation;
- \* coarsened substrate;
- \* channel pattern shift from meandering to straight;
- \* decreased bank stability;
- \* increased channel profile uniformity;
- \* increased water velocity;
- \* decreased water depth;
- \* decreased wetted surface area;
- \* decreased depth and velocity variability;
- \* altered stage-discharge relationships;
- \* lowered water tables in streamside areas;

Wesche further links these hydraulic and morphologic changes to differences in stream habitat for fish and other aquatic life.

These same hydraulic and morphologic changes occur whether a stream is confined to a straight course by a highway, steep hill slope, alluvial fan, high stream terrace or any other man-made or natural feature that confines the sinuosity of the stream. In the western United States where many river valleys are flanked by mountain slopes, it is common for the some part of the river to be confined to a straight course by natural features. Streambanks confined by natural features often resemble streambanks confined by man-made features in being unstable and sparsely vegetated. Talus on steep residual slopes resembles rip-rip. High, unstable streambanks

adjacent to alluvial fans are typically eroding. Stream sinuosity is typically low where banks are confined, whether by natural or man-made features.

The straight state was identified in Reach 1 of Rock Creek and in Reach 4 and 5 of the BHR. Confinement and sinuosity of straight reaches were evaluated from 1:24,000 scale USGS quads. Results are discussed with respect to each reach:

**Reach 1 Rock Creek:** The left bank (facing downstream) of the portion of Rock Creek classified as straight is confined by a residual mountain slope and alluvial fans along 77 percent (4.1 miles) of its length. The remaining 23 percent (1.2 miles) is not confined. The sinuosity of this reach is very low (1.04).

**Reach 4 Big Hole River:** About 39 percent (0.9 miles) is confined on both banks by alluvial fans. About 15 percent (0.4 miles) is confined by an alluvial fan along the left bank. The reach is confined by residual mountain slope along 28 percent (0.7 miles) of the left bank. The remaining 18 percent (0.4 miles) of Reach 4 is not confined. The sinuosity of this reach is very low (1.08)

**Reach 5 Big Hole:** About 38 percent (0.5 miles) of this reach is confined by an alluvial fan along the right bank. The remaining 62 percent (0.8 miles) is not confined. The sinuosity of this reach is very low (1.02).

The sinuosity of straight reference reaches is similar to channelized reaches of the CFR. The degree of confinement of Reach 1 Rock Creek and Reach 4 Big Hole are also similar to that of channelized reaches of the CFR.

## 2.4 Ecoregion

On page D2-5 Reiser criticizes the use of Ecoregion as the broadest level of the hierarchical classification. The following is excerpted from Dr. Reiser's report.

"The State used ecoregion as the broadest category in its classification system. The use of ecoregion as originally intended by Omernik (1987) was based more on large-scale land attributes and terrestrial habitats and communities (Omernik 1987). The landform of the ecoregion containing the majority of the CFR (*Clark Fork River*), plus the Big Hole, Beaverhead, and Ruby rivers (the Montana Valley and Foothill Prairies Ecoregion) was so diverse that Omernik (1987) could not discern any appropriate pattern from his component maps and therefore a variety of landforms were lumped into one. Omernik (1987) did hypothesize that his ecoregions and their components display regional patterns that are reflected in spatially variable combinations of causal factors, including climate, mineral availability (soils and geology), vegetation, and physiography. However, he cited two references that stressed that ecoregion maps are products of hypotheses that must be tested and improved. This was not done by the State."

The abstract of the above cited report (Omernik 1987) says in part:

"A map of ecoregions of the conterminous United States has been compiled to assist managers of aquatic and terrestrial resources in understanding the regional patterns of the realistically attainable quality of these resources. The ecoregions are based on perceived patterns of a combination of causal and integrative factors including land use, land surface form, potential natural vegetation and soils. .... they center on aquatic ecosystems - mainly attainable ranges in chemical quality, biotic assemblages, and lake trophic state.

Omernik states the primary function of the ecoregion map is to provide a framework to: 1) compare similarities and differences of land/water relationships; 2) establish water quality standards that are in tune with regional patterns of tolerance and resilience to human impacts; 3) locate monitoring, demonstration, or reference sites; 4) extrapolate from existing, site-specific studies; and 5) predict the effects of changes in land use and pollution controls. Use of Ecoregions as the broadest level of the hierarchical classification is appropriate.



### 3.0 LITERATURE CITED

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- Reiser, D.W. 1995. Expert Report - United States District Court, District of Montana, Helena Division. State of Montana versus Atlantic Richfield Co. (No. CF-83-317-HLN-PGH).
- Wesche, T.A. 1995. Expert Report - United States District Court, District of Montana, Helena Division. State of Montana versus Atlantic Richfield Co. (No. CF-83-317-HLN-PGH).

**APPENDIX A**  
**LAND USES BY ECOREGION, CFR AND REFERENCE BASINS**



Table A-1. Land uses by Ecoregion, CFR and reference basins.

BASIN	ECOREGION	LAND USE	-----AREA-----	
			(mi <sup>2</sup> )	(%)
Clark Fork Subbasin	N. Rockies	Urban	2.6	0.3
Clark Fork Subbasin	N. Rockies	Agriculture	7.7	0.9
Clark Fork Subbasin	N. Rockies	Range	177.1	20.7
Clark Fork Subbasin	N. Rockies	Forest	645.7	75.3
Clark Fork Subbasin	N. Rockies	Water	1.0	0.1
Clark Fork Subbasin	N. Rockies	Wetland	0.0	0.0
Clark Fork Subbasin	N. Rockies	Barren	19.4	2.3
Clark Fork Subbasin	N. Rockies	Tundra	3.8	0.4
Clark Fork Subbasin	N. Rockies	Total	857.2	100.0
Rock Creek	N. Rockies	Urban	0.2	0.0
Rock Creek	N. Rockies	Agriculture	2.3	0.3
Rock Creek	N. Rockies	Range	70.6	9.1
Rock Creek	N. Rockies	Forest	691.0	89.0
Rock Creek	N. Rockies	Water	1.4	0.2
Rock Creek	N. Rockies	Wetland	3.2	0.4
Rock Creek	N. Rockies	Barren	7.5	1.0
Rock Creek	N. Rockies	Tundra	0.5	0.1
Rock Creek	N. Rockies	Total	776.6	100.0
Flint Creek	N. Rockies	Urban	1.2	0.6
Flint Creek	N. Rockies	Agriculture	10.0	4.4
Flint Creek	N. Rockies	Range	23.7	10.5
Flint Creek	N. Rockies	Forest	183.8	81.4
Flint Creek	N. Rockies	Water	4.6	2.0
Flint Creek	N. Rockies	Wetland	0.2	0.1
Flint Creek	N. Rockies	Barren	2.3	1.0
Flint Creek	N. Rockies	Tundra	0.0	0.0
Flint Creek	N. Rockies	Total	225.8	100.0
Silver Bow	N. Rockies	Urban	2.7	1.4
Silver Bow	N. Rockies	Agriculture	0.0	0.0
Silver Bow	N. Rockies	Range	39.2	20.5
Silver Bow	N. Rockies	Forest	139.0	72.5
Silver Bow	N. Rockies	Water	0.0	0.0
Silver Bow	N. Rockies	Wetland	0.0	0.0
Silver Bow	N. Rockies	Barren	9.1	4.7
Silver Bow	N. Rockies	Tundra	1.7	0.9
Silver Bow	N. Rockies	Total	191.7	100.0

Table A-1. Continued.

BASIN	ECOREGION	LAND USE	-----AREA----- (mi <sup>2</sup> )	(%)
CLARK FORK ALL	N. Rockies	Urban	6.7	0.3
CLARK FORK ALL	N. Rockies	Agriculture	20.0	1.0
CLARK FORK ALL	N. Rockies	Range	310.5	15.1
CLARK FORK ALL	N. Rockies	Forest	1659.5	80.9
CLARK FORK ALL	N. Rockies	Water	7.0	0.3
CLARK FORK ALL	N. Rockies	Wetland	3.4	0.2
CLARK FORK ALL	N. Rockies	Barren	38.3	1.9
CLARK FORK ALL	N. Rockies	Tundra	6.0	0.3
CLARK FORK ALL	N. Rockies	Total	2051.3	100.0
Clark Fork Subbasin	MVFP	Urban	13.6	1.4
Clark Fork Subbasin	MVFP	Agriculture	148.9	15.5
Clark Fork Subbasin	MVFP	Range	470.3	49.0
Clark Fork Subbasin	MVFP	Forest	297.2	30.9
Clark Fork Subbasin	MVFP	Water	0.7	0.1
Clark Fork Subbasin	MVFP	Wetland	12.9	1.3
Clark Fork Subbasin	MVFP	Barren	16.8	1.7
Clark Fork Subbasin	MVFP	Tundra	0.0	0.0
Clark Fork Subbasin	MVFP	Total	960.4	100.0
Rock Creek	MVFP	Urban	0.0	0.0
Rock Creek	MVFP	Agriculture	3.8	3.4
Rock Creek	MVFP	Range	73.1	65.4
Rock Creek	MVFP	Forest	30.0	26.8
Rock Creek	MVFP	Water	0.0	0.0
Rock Creek	MVFP	Wetland	4.9	4.4
Rock Creek	MVFP	Barren	0.0	0.0
Rock Creek	MVFP	Tundra	0.0	0.0
Rock Creek	MVFP	Total	111.7	100.0
Flint Creek	MVFP	Urban	0.5	0.2
Flint Creek	MVFP	Agriculture	51.2	18.7
Flint Creek	MVFP	Range	124.3	45.5
Flint Creek	MVFP	Forest	93.0	34.0
Flint Creek	MVFP	Water	0.3	0.1
Flint Creek	MVFP	Wetland	0.0	0.0
Flint Creek	MVFP	Barren	3.9	1.4
Flint Creek	MVFP	Tundra	0.0	0.0
Flint Creek	MVFP	Total	273.2	100.0

Table A-1. Continued.

BASIN	ECOREGION	LAND USE	-----AREA----- (mi <sup>2</sup> )	(%)
Silver Bow	MVFP	Urban	14.7	5.2
Silver Bow	MVFP	Agriculture	10.4	3.7
Silver Bow	MVFP	Range	177.6	62.9
Silver Bow	MVFP	Forest	62.2	22.0
Silver Bow	MVFP	Water	0.0	0.0
Silver Bow	MVFP	Wetland	0.0	0.0
Silver Bow	MVFP	Barren	17.5	6.2
Silver Bow	MVFP	Tundra	0.0	0.0
Silver Bow	MVFP	Total	282.5	100.0
CLARK FORK ALL	MVFP	Urban	28.8	1.8
CLARK FORK ALL	MVFP	Agriculture	214.3	13.2
CLARK FORK ALL	MVFP	Range	845.3	51.9
CLARK FORK ALL	MVFP	Forest	482.3	29.6
CLARK FORK ALL	MVFP	Water	1.0	0.1
CLARK FORK ALL	MVFP	Wetland	17.8	1.1
CLARK FORK ALL	MVFP	Barren	38.2	2.3
CLARK FORK ALL	MVFP	Tundra	0.0	0.0
CLARK FORK ALL	MVFP	Total	1627.8	100.0
Big Hole	MVFP	Urban	3.4	0.3
Big Hole	MVFP	Agriculture	102.1	8.3
Big Hole	MVFP	Range	724.0	59.1
Big Hole	MVFP	Forest	335.8	27.4
Big Hole	MVFP	Water	0.3	0.0
Big Hole	MVFP	Wetland	58.9	4.8
Big Hole	MVFP	Barren	0.8	0.1
Big Hole	MVFP	Tundra	0.0	0.0
Big Hole	MVFP	Total	1225.3	100.0
Beaverhead	MVFP	Urban	6.8	0.3
Beaverhead	MVFP	Agriculture	326.9	12.1
Beaverhead	MVFP	Range	2099.9	77.6
Beaverhead	MVFP	Forest	188.0	6.9
Beaverhead	MVFP	Water	19.0	0.7
Beaverhead	MVFP	Wetland	63.2	2.3
Beaverhead	MVFP	Barren	1.1	0.0
Beaverhead	MVFP	Tundra	0.0	0.0
Beaverhead	MVFP	Total	2704.9	100.0

Table A-1. Continued.

BASIN	ECOREGION	LAND USE	-----AREA----- (mi <sup>2</sup> )	(%)
Ruby River	MVFP	Urban	0.7	0.2
Ruby River	MVFP	Agriculture	82.3	22.0
Ruby River	MVFP	Range	204.4	54.7
Ruby River	MVFP	Forest	78.8	21.1
Ruby River	MVFP	Water	1.6	0.4
Ruby River	MVFP	Wetland	3.6	1.0
Ruby River	MVFP	Barren	2.3	0.6
Ruby River	MVFP	Tundra	0.3	0.1
Ruby River	MVFP	Total	374.0	100.0





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**APPENDIX G**  
**RESPONSE TO THOMAS C. GINN'S EXPERT REPORT ON SURFACE**  
**WATER RESOURCES, PAGE 19, POINT NO. 2**

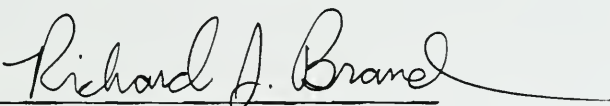




**Response to Thomas C. Ginn's Expert Report on Surface Water  
Resources, Page 19, Point No. 2**

**By Dr. Richard J. Brand**

**October 20, 1995**

  
Richard J. Brand



**Response to Thomas C. Ginn's Expert Report on Surface Water Resources, Page 19, Point No. 2: EPA vs. Montana (MT) Total Recoverable Analysis of Metals.**

ARCO's consultant Dr. Thomas C. Ginn has performed an analysis of the data collected by the State for comparison of the EPA vs. the MT methods for recovering total metal content of water samples. ARCO has emphasized the use of t-tests, both using and excluding data that were at detection limits. These tests were applied to the paired differences between results obtained by making measurements using both the EPA and MT total recovery methods on each of a collection of water samples (see Tables A-4, A-5 and A-6 in Montana's January 1995 aquatics report).

The State originally used the sign test to assess the statistical significance of the difference between the EPA and MT methods. The sign test was chosen because it is more appropriate for the type of data distributions that are found with the EPA and MT comparison data. The sign test is a "distribution-free" test which does not require that the distribution of the paired differences be normally distributed. The sign test uses a sensible strategy for dealing with tied pairs, which give a difference equal to zero. The sign test allocates one half of the ties in favor of the EPA method and one half in favor of the MT method. In this way, the sign test is designed to function reasonably and fairly even if there is a substantial fraction of the paired differences that are equal to zero.

By contrast, the t-test is designed to be utilized when there is a substantially normal distribution of paired differences. The substantially non-normal distributions of partial differences encountered in this case invalidate the t-test. In summary, Dr. Ginn's criticism of the State's analysis is not well-taken because it is based on the t-test which is an inappropriate statistical test in this case.

An alternative "distribution-free" test that could have been used by the State is the Wilcoxon Signed Ranks Test. It is known to be statistically more efficient than the rather conservative sign test. When the signed rank test (SRT) is used it confirms the results of the sign test. The two-sided p-values for the SRT for cadmium, copper, lead, and zinc EPA vs. MT paired differences are 0.1384, 0.0001, 0.0029, and 0.0001, respectively. By comparison, the corresponding two-sided p-values for the sign test are 0.441, 0.0002, 0.0436, and 0.0001 respectively. Thus it can be concluded that the observed differences between the EPA and MT results for copper, lead, and zinc are unlikely to be due to chance alone.

Moreover, this can be concluded even when using two-sided tests which allow for the possibility that the MT method could lead to higher values than the EPA method. It is a known fact that the EPA digestion method is more intense and should recover more of the



metal and, in fact, there is past evidence to this effect. This basic knowledge could have justified a one-sided test which would have given lower, more significant p-values. (For example all of the sign-test p-values would have been cut in half.)

It is important to examine the statistical significance test results in connection with the corresponding descriptive findings.

For cadmium, 36/42 paired measurements were the same and led to a difference of zero, primarily because so many of the samples were at detection limits for both methods. Of the remaining six measurements, all showed higher findings by the EPA method. In this situation, the data sample did not contain very much information for comparing the two methods and the resulting two-sided p-value, as noted above, was relatively large. Thus, these data do not demonstrate that the EPA method gives higher values; however, it cannot be concluded that the EPA does not give higher values.

For copper, 14/45 paired measurements were the same. Of the remaining 31, 28 gave higher values for the EPA method while three gave higher values for the MT method. The difference was highly significant by the appropriate SRT and sign tests.

For lead, 24/42 paired measurements were the same. Of the remaining 18, 16 gave higher values for the EPA method while only two gave higher values for the MT method. These observed differences were again statistically significant at a noteworthy level, particularly by the more efficient SRT method.

For zinc, only 4/42 measurements were the same so, among the four metals, this metal gave the best opportunity for a sensitive statistical assessment of the difference between the two methods. Of the remaining 37 paired differences, 33 gave higher values for the EPA method, while only four pairs gave a higher value for the MT method. The observed differences were highly statistically significant by SRT and sign tests.

Taken overall it appears reasonable to conclude that the EPA method for recovering total metal content of the water samples gives higher values. Conversely, it appears reasonable to conclude that the MT method used by the State for its water sampling, does not extract total metal as thoroughly as the EPA method and, therefore, the MT method tends to underestimate the actual total metal concentration in the water samples.









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**APPENDIX H**  
**COMMENTS ON ISSUES RELATED TO THE USE OF DISSOLVED**  
**METAL VS TOTAL RECOVERABLE METAL IN SETTING AND**  
**DETERMINING COMPLIANCE WITH MONTANA WATER QUALITY**  
**STANDARDS**

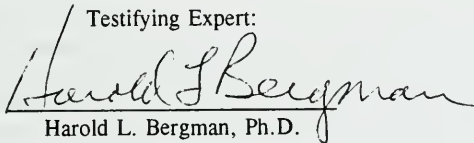


COMMENTS ON ISSUES RELATED TO THE USE OF  
"DISSOLVED METAL" vs. "TOTAL RECOVERABLE METAL"  
IN SETTING AND DETERMINING COMPLIANCE WITH  
MONTANA WATER QUALITY STANDARDS

Prepared by:

Harold L. Bergman

Testifying Expert:

A handwritten signature in cursive script, reading "Harold L. Bergman". The signature is written in dark ink and is positioned above the printed name and affiliation.

Harold L. Bergman, Ph.D.  
Department of Zoology and Physiology  
University of Wyoming  
Laramie, Wyoming 82071

Date:

October 12, 1995



The purposes of water quality criteria (WQC) and standards for protection of aquatic life are to limit the concentration, duration and frequency of exposure to toxic chemicals, so as to prevent adverse acute or chronic effects on aquatic organisms (U.S. EPA 1987). The criteria and standards for most priority pollutants, including the toxic metals, are based on extensive databases of results from acute and chronic toxicity tests using a wide variety of aquatic organisms including fish, aquatic invertebrates and plants. Many but not all of these toxicity tests were conducted in the laboratory with relatively clean waters having relatively low concentrations of complexing chemical constituents. Because of this, the EPA originally considered the use of "dissolved metal" concentrations but instead recommended the use of "total recoverable metal" concentrations for setting and determining compliance with water quality standards in fresh water.

### **Recent Guidance from U.S. EPA on use of "Dissolved Metal" and "Total Recoverable Metal" Concentrations in Water Quality Standards**

In 1993, the U.S. EPA released new guidance that states "the use of dissolved metal to set and measure compliance with water quality standards is the recommended approach, because dissolved metal more closely approximates the bioavailable fraction of metal in the water column than does total recoverable metal" (Prothro, 1993). However, in the same memorandum (Prothro 1993) the U.S. EPA qualifies this recommendation, as follows:

"The reasons for the potential consideration of total recoverable measurements include risk management considerations not covered by evaluation of water column toxicity. The ambient water quality criteria are neither designed nor intended to protect sediments, or to prevent effects due to food webs containing sediment dwelling organisms. A risk manager, however, may consider sediments and food chain effects and may decide to take a conservative approach for metals, considering that metals are very persistent chemicals. This conservative approach could include the use of total recoverable metal in water quality standards."

The particular relevance and appropriateness of such risk management considerations (i.e., to use total recoverable metal to protect aquatic biota from sediment and food chain routes of exposure) were also emphasized in further recommendations and guidance from the U.S. EPA that were specific to the State of Montana (Wuerthele 1995).

### **Recent Research Findings Relevant to the Risk Management Considerations Recommended by the U.S. EPA for Establishing Water Quality Standards**

Recent research findings for the Clark Fork River in Montana relate specifically to the risk management considerations mentioned by the U.S. EPA in its recent guidance (Prothro 1993, Wuerthele 1995) and support the use of total recoverable metals in Montana's water quality standards. One additional risk management consideration, mentioned in the U.S. EPA guidance from Wuerthele (1995), also applies to the prevailing conditions in the Clark Fork River and further supports Montana's use of total recoverable metals for water quality standards. These research findings are as follows:



- The bed sediments of the Clark Fork River in Montana are heavily contaminated with metals released from past mining and milling operations (e.g., Moore and Luoma 1990).
- Food route exposure to metals and resulting injuries to fish populations in the Clark Fork River have been documented by several studies published by Woodward et al. (1994, 1995).
- Temporal and spacial variability in ambient water quality conditions on the Clark Fork River, such as the documented acid pulse events that have resulted in fish kills (Lipton et al. 1995), can greatly increase the adverse effects of water column metals on fish and other aquatic biota. Thus, metals that may be sorbed to particulates in the water column at one point in the river (and therefore not measured if a "dissolved metal" determination were used) will desorb from the particulates or otherwise become more "bioavailable" when water quality conditions change downriver (such as occurs when pH, hardness and alkalinity decrease).

### **Use of "Montana Total Recoverable Metal" vs. "EPA Total Recoverable Metal" Concentrations for Montana Water Quality Standards**

Analytical determinations and State Water Quality Standards based on "Montana Total Recoverable Metal" are less stringent than "EPA Total Recoverable Metal." The Montana method involves acidification of a water sample in the field, followed by settling and decanting of the water sample, and metal analysis of the decanted water. The EPA method, on the other hand, includes an additional step, specifically the digestion of the water sample in hot mineral acid, which will tend to release more of the metal from particulate matter and yield a higher measured metal concentration.

Given that the U.S. EPA advises that "total recoverable metal" (the U.S. EPA is referring, here, to the "EPA total recoverable metal" method) is justified when risk management considerations warrant, the State of Montana would be justified in using the "EPA total recoverable metal" method rather than the less stringent Montana method. Moreover, since all three risk management considerations, listed above, are known to apply to the Clark Fork River, and are not merely suspected risk factors, the State of Montana should switch to the "EPA total recoverable metal" method, at least for water quality standards on the Clark Fork River.

### **Conclusions and Recommendations**

- The use of "dissolved metal" concentrations for water quality standards would be inappropriate for the State of Montana, particularly in water bodies such as the Clark Fork River, which have extremely high sediment metal concentrations due to past metal mining and smelting operations in the watershed.
- Risk management considerations, related to the protection of aquatic biota from metal contaminated sediments, from food-chain exposure routes to fish, and from





temporal and spacial variation in water quality conditions that can increase toxicity of metals downstream, dictate that Montana should use "total recoverable metal" concentrations in setting and determining compliance with water quality standards.

- Because of the risk management considerations listed above for the Clark Fork River, and because the "Montana total recoverable metal" method is less stringent than the "EPA total recoverable metal" method, the State of Montana would be justified and should use the "EPA total recoverable metal" method to enforce water quality standards on the Clark Fork River.

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**APPENDIX I**  
**A COMPARISON OF METALS DIGESTION BY CUTTHROAT TROUT**  
**BETWEEN THREE DIETS CONTAMINATED UNDER NATURAL**  
**CONDITIONS AND A FORTH DIET CONTAMINATED UNDER**  
**LABORATORY CONDITIONS**



Distribution of Metals During Digestion by Cutthroat Trout  
Fed Invertebrate Diets Contaminated in the Clark Fork River  
or under Laboratory Conditions

Prepared by:

Daniel F. Woodward

Testifying Expert:

A handwritten signature in cursive script, reading "Daniel F. Woodward", written over a horizontal line.

Daniel F. Woodward  
National Biological Service  
Midwest Science Center  
Jackson, Wyoming 83001

Date:

October 17, 1995





Distribution of Metals During Digestion by Cutthroat Trout  
Fed Invertebrate Diets Contaminated in the Clark Fork River  
or under Laboratory Conditions

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Submitted by:

Daniel F. Woodward  
National Biological Service  
Midwest Science Center  
Jackson Field Station  
Jackson, Wyoming 83001

Harold L. Bergman  
University of Wyoming  
Department of Zoology and Physiology  
Laramie, Wyoming 82071

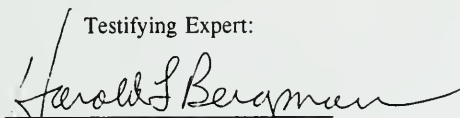
Contributing authors:

Aida Farag - National Biological Service  
Joseph Meyer - University of Wyoming  
Mike Suedkamp - University of Wyoming



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Testifying Expert:

A handwritten signature in cursive script, reading "Harold L. Bergman". The signature is written in dark ink and is positioned above a horizontal line.

Harold L. Bergman, Ph.D.  
Department of Zoology and Physiology  
University of Wyoming  
Laramie, Wyoming 82071

Date:

October 17, 1995



The waters, sediments and biota of the Clark Fork River (CFR), Montana, have elevated concentrations of arsenic (As), copper (Cu), cadmium (Cd), lead (Pb) and zinc (Zn) (Moore and Luoma 1990). Metals are available to organisms in the CFR through uptake across the gill and across the gut from the diet. Benthic invertebrates are important as food sources for fish and waterfowl, and they are essential in trophic energy transfer and nutrient cycling. Thus, fish and other vertebrates feeding at higher trophic levels may be chronically exposed to metals through the food chain as well as the water.

Rainbow trout (*Oncorhynchus mykiss*) and brown trout (*Salmo trutta*) fed diets of benthic macroinvertebrates from the CFR, died or exhibited reduced growth. These effects were associated with elevated concentrations of metals in whole body samples (Woodward et al. 1994, Woodward et al. 1995). Physiological and biochemical changes (e.g., decreased growth, lipid peroxidation, and histological changes) have been measured in fish from laboratory experiments and/or in fish collected from the field where metals concentrations were also elevated in whole body or specific tissues (Farag et al. 1994, Farag et al. 1995, Woodward et al. 1995). Therefore, dietary metals exposure, physiological impact, and metals accumulation are linked.

The specific forms of metal-organic complexes in invertebrates consumed by fish may affect the bioavailability and thus the toxicity of those metals to fish. Metals in ionic forms and loosely associated with organics are less toxic to fish via the diet than metals covalently bound to, or tightly complexed with organics in the diet (Hodson 1988, Harrison and Curtis 1992). Quantitatively, metal concentrations may be similar in the tissues of fish fed diets containing metals in ionic forms and in fish fed diets where metals are bound to organics. However, the qualitative aspects of the two diets may be quite different. The metal-organic complexes in natural diets increase the bioavailability of the metals to the fish and may be more



toxic (Hodson 1988) than diets in an ionic form.

Results of research by Mount et al. (1994) on metals contamination in the CFR gives further evidence that metals in the ionic form are not toxic. They observed few adverse effects on rainbow trout when they were fed brine shrimp (*Artemia* sp.) that had received an aqueous exposure to metals (Mount et al. 1994). Brine shrimp were exposed for less than 24 h following hatching and were not fed before harvesting and being fed to fish. Therefore, it is doubtful that normal metabolic assimilation occurred in the newly hatched brine shrimp; as a result, the metals attached to brine shrimp were likely in the free ionic form.

The brine shrimp prepared in the laboratory were exposed to metals exclusively via the water; however, natural invertebrates in the CFR are exposed to metals via water, diet and sediment. The exposure of natural populations is continuous and metabolism in individuals of those populations occurs as the individuals develop and grow. The impacts of metabolized metals far outweigh that of simple ions (Hodson 1988) in the food-chain, and it is known that As, Cd and Cu are metabolized by aquatic biota (Craig 1986). We believe the metals present in the *Artemia* diet (Mount et al. 1994) were present in different form(s) than metals present in the macroinvertebrate diets collected from the CFR. For example, metals may be adsorbed to *Artemia* but may not be complexed with organic biomolecules such as proteins and/or amino acids as in the benthic macroinvertebrates.

The digestive processes involved when fish are fed these various diet types may differ. We hypothesize that metals naturally incorporated into invertebrates collected from the CFR are processed differently during digestion by cutthroat trout, than diets made up of *Artemia* exposed to metals in the laboratory for 24 h.





## METHODS

Four dietary treatments were fed to cutthroat trout in the laboratory. Fish received three dietary treatments of invertebrates collected from naturally contaminated sources in the CFR. These three diets were collected near Turah Bridge (TB), Gold Creek (GC) and below Warm Springs (WS) ponds, as reported in Woodward et al. (1995). The fourth dietary treatment consisted of *Artemia* exposed in the laboratory for 18-24 hours after hatching in a mixture of metals in water, as reported in Mount et al. (1994).

### Experimental Design

Cutthroat trout of approximately 100 g each were transferred from the Jackson National Fish Hatchery to the Jackson Field Station 15 days before the experiment. Trout were anesthetized 5 days later with MS222, marked with brands cooled in liquid nitrogen, and randomly distributed to achieve 12 fish in each of five, 300-L experimental chambers (flow rate was 6.5 - 8.0 L/min., or 34 volume additions per day). Four unique brands identified the different dietary treatments and three unique locations of the brands identified three experimental replicates within a chamber. Therefore, all diets were represented in each of the experimental chambers. The experimental unit was the chamber, thus  $N = 5$ , and the digestive fluids from replicate fish within a chamber were combined for chemical analyses. Fish were fed a commercial trout food until 48 hr prior to the beginning of the experiment.

### Diet Preparation

The invertebrate diets from the CFR were the same as those used by Woodward et al. (1995). For experiments performed by Woodward et al. (1995), the raw invertebrates were pasteurized and prepared into dry, pelletized diets at the U.S. FWS, Fish Technology Center, Bozeman, Montana. The diet was processed to eliminate disease potential from the food organisms and to assure the proper vitamins and minerals were present in the diets.

The *Artemia* diets were prepared at the Jackson Field Station



according to procedures reported by ENSR (1994) and Mount et al. (1994). Six grams of *Artemia* cysts (Argentemia, Gold Label, Lot #BG1204J) were hatched in 1 L of deionized water with 20 g of Instant Ocean sea salt and a mixture of As, Cd, Cu, Pb, and Zn. The solution was aerated and maintained at 28°C for 48 h, after which the *Artemia* were removed and allowed to depurate in moderately hard reconstituted water for 5 min.

The concentrations of four essential amino acids -- valine, phenylalanine, leucine, and isoleucine -- were measured on five samples of each diets. The presence of interfering compounds (e.g., glucosamine, from acid digestion of chitin in the invertebrates) in the diets made the measurements of additional essential amino acids impossible. The concentrations of essential amino acids in the TB, GC, and WS diets were similar to each other; but in all cases, were about 25% to 75% less than the *Artemia* (Table 1).

#### Exposure

All diets were stirred with de-ionized water to form a paste that could be dispensed through animal feeding needles (Popper and Sons, Inc., #9915). Three-inch, 15-gauge feeding needles were attached to syringes and 1/2 ml of diet was dispensed into the digestive tract of fish anesthetized with MS222. Fish were observed for approximately 30 seconds following each feeding and the regurgitation of any food was noted. If a small amount of food appeared in the water surrounding the fish's mouth, it was noted that slight regurgitation occurred and the fish was not re-fed. If food pulsed into the water with force or a greater amount of food was released, we designated this as extensive regurgitation and re-fed the fish. Fish were fed for three consecutive days and sacrificed on the fourth day, approximately 16 hr after the last feeding.

Each fish was sacrificed with an overdose of MS222 and dissected immediately. The digestive tract was divided into 3 sections by ligation: stomach, anterior intestine with pyloric caeca, and hind gut. The bile duct was also ligated. Then the



entire digestive tract was removed, placed into a cooled petri dish, and rinsed with refrigerated physiological saline. The anterior intestine was injected with 0.5 ml of physiological saline, and the anterior intestine and the pyloric caecae were massaged gently with forceps. Then, the junction between the anterior intestine and the hind gut was severed, allowing the contents of the anterior intestine and the pyloric caecae to flow into a cryo-vial. This process was repeated a second time and the total sample of intestinal contents was immediately frozen in liquid nitrogen. Approximately 25 min. elapsed from the time each fish was removed from the water until the complete sample of intestinal contents was frozen. Anterior intestine with pyloric caecae, hind gut and feces were also collected and frozen in liquid nitrogen.

All intestinal content samples were transported on dry ice to the Fish Physiology and Toxicology Laboratory, Laramie, Wyoming, where they were stored at -82°C until analyses were performed. In addition, subsamples of the five diets were transported to the Fish Physiology and Toxicology Laboratory and stored at -20°C until metal analyses were performed. The pyloric caecae were dissected away from the anterior intestine, and anterior intestine, pyloric caecae, hind gut and feces are currently being stored at -82°C.

#### Component Separation and Metal Analyses

Two separations were performed on the intestinal contents and the concentrations of As, Cd, Cu, and Pb were measured in each four components (Figure 1). During the first separation, samples were centrifuged at 5,000 g for 20 minutes to separate the intestinal fluids from the fat and particulates. The supernatant (intestinal fluid) was removed with an acid-washed pipette and centrifuged further at 3,500 g, through a Millipore 10,000 nominal molecular-weight cutoff, polysulfone micropartition tube. Proteins (retentate) remained on the membrane in the process and amino acids and free metal ions collected in the filtrate. There was a four component



separation: (1) fat and particles, (2) intestinal fluids, (3) proteins, (4) amino acid/free metal ions. Arsenic, Cd, Cu and Pb were measured in each component with atomic absorption spectroscopy (AAS) using a Varian SpectraAA-600 equipped with a graphite furnace and Zeeman background correction. Total protein was measured with a Biorad (undated) protein assay, in the intestinal fluid and in the amino acid/free metal ion fraction of the intestinal fluid. Protein measurements in the latter fraction documented any movement of protein through the microfilters.

A procedure utilizing high performance liquid chromatography (HPLC) to separate amino acids from free metals was outlined in the protocol (Figure 1). We would have collected the amino acid/free metal fractions from the HPLC and measured the concentrations of metals in these fractions with AAS. However, samples must first be prepared with a series of buffers and a chemically harsh derivitization before they are injected into an HPLC. We were concerned that the amino acids would undergo conformational changes during the HPLC preparation steps. Therefore, deviations from the procedure outlined in the protocol are being explored to improve our ability to separate amino acids from the free metals and, at the same time, keep the amino acids intact. Additionally, we need to document the presence and quantity of amino acids in this amino acid/free metal ion fraction in much the same way we defined the presence of proteins in the protein fraction. If the concentrations of amino acids in the amino acid/free metal ion fraction are negligible, there would be no gain by attempting to separate the two. However, due to technical limitations, we have been unable to complete this task.

### Statistics

The concentration of each metal in the various separation fractions was expressed as a percent of the total concentration of that metal, in order to standardize the concentrations of metals in the intestinal contents. For example, the amount of Cu





in the particulate and fluid fractions are expressed as a percentage of the total Cu measured as  $\mu\text{g/g}$ , for that fractionation process. The amounts of Cu in the protein and amino acid/free ion fractions are expressed as a percentage of the total Cu measured for that fractionation process. After the Levene's test was performed on the percentage values to confirm that the homogeneity assumption for ANOVA had been met, one-way ANOVAs followed by Tukey multiple means comparisons were performed on percent values for each metal.

## RESULTS AND DISCUSSION

The feeding procedure was successful and we noted feces in the experimental chambers throughout the 3-day exposure. There were five instances where fish regurgitated food to the extent that they had to be fed a second time. All five of these instances occurred when fish were fed the diet of *Artemia* exposed to metals. Therefore, trout were unwilling to accept the diet of *Artemia* exposed to metals in 11% of the feedings. There was a slight regurgitation once when a trout was fed the diet from GC and twice when trout were fed the diet from WS. However, these instances of regurgitation were slight, and fish were not fed a second time.

Texture, quantity, and appearance of the intestinal contents differed for trout between diet treatments. A smaller amount of digestive fluid was collected from all fish fed the diet prepared with *Artemia*, and the fluids were all translucent with red hues when compared to the digestive fluids from fish fed the invertebrate diets collected from the CFR. The fat in the intestinal contents of fish fed diets of *Artemia* was spherical, orange in color, and would break easily when touched. However, the fat in the intestinal contents of fish fed invertebrates collected from the CFR was disc-shaped, white in color, and remained on the surface of the samples. Thus, not only did the *Artemia* diet seem slightly less palatable, but the digestion of these two diet types affected the physical characteristics of fat



in the intestinal contents differently.

#### Copper

The mean concentration of Cu in the *Artemia* diet was 106  $\mu\text{g/g}$  (Table 2), and was nearest to the concentration of Cu present in invertebrates from TB (105  $\mu\text{g/g}$ ), which was the reference diet in Woodward et al. (1995). Our goal was to achieve a Cu concentration in the *Artemia* diet which was more similar to that in the test diets, WS and GC (191 and 188  $\mu\text{g/g}$ , respectively).

Copper was partitioned differently in the intestinal contents of fish fed *Artemia* diets when compared to fish fed diets collected from the natural source in the CFR. Trout fed *Artemia* had a significantly greater percent of Cu in the intestinal fluid (66%) and a corresponding smaller percent of Cu in the particulates (34%) than trout fed invertebrates from GC (44% and 56%, respectively) and WS (48% and 52%, respectively) (Table 2). The percentage of Cu in the intestinal fluid from fish fed the TB diet (54%) was also less than the percentage of Cu in fish fed the *Artemia* diet (66%), but this difference was not significant.

The percent of Cu in the amino acid/free ion fraction of the intestinal fluids was 79% for trout fed the *Artemia* diet, which was greater than the natural diets, TB 69%, GC 72%, and WS 72% (Table 3). While there was a trend of higher Cu in the intestinal fluids of fish fed the *Artemia*, the difference was only significant between fish fed the TB diet and fish fed the *Artemia* diet.

#### Lead

Lead in the *Artemia* diet was 23  $\mu\text{g/g}$  and was greater than the concentrations of all three natural diets (GC, 18  $\mu\text{g/g}$ ; WS, 17  $\mu\text{g/g}$ ; and TB 8  $\mu\text{g/g}$ ) (Table 4). Lead was also partitioned differently in the intestinal contents of fish fed different types of diets. The percent of Pb in the intestinal fluid was statistically greater in fish fed *Artemia* (29%) than fish fed any of the three natural diets collected from the CFR (TB, 17%; GC,



12%; and WS, 10%). There were no statistical differences between the amount of Pb in protein versus the amino acid/free metal ion fractions of the intestinal fluids.

#### Arsenic and Cadmium

The *Artemia* diet contained 40  $\mu\text{g/g}$  As and 10  $\mu\text{g/g}$  Cd, both of which are greater than the measured As and Cd in the natural diets (TB, 5 and 2  $\mu\text{g/g}$ ; GC, 21 and 2  $\mu\text{g/g}$ ; and WS, 19  $\mu\text{g/g}$  and 2  $\mu\text{g/g}$ , respectively). The percentages of As and Cd were higher in the intestinal fluids of fish fed the *Artemia* diets as compared to the intestinal fluids of fish fed the diets collected from the CFR, but these differences were not significant (Tables 5 and 6). For fish fed the *Artemia* diet, the percentage of As and Cd in the intestinal fluid was 56% and 60%, respectively; versus 41 to 50% As and 38% to 48% Cd in the intestinal fluid of fish fed the natural diets.

#### Summary

The significant differences observed in the intestinal contents were between the *Artemia* diet and one or more of the CFR diets and occurred for the partitioning of Cu and Pb. There were no statistical differences in the percentage distribution of As, Cd, Cu, or Pb in the intestinal contents of fish fed the three diets collected from the CFR. The TB, GC and WS diets were also similar to each other in terms of the concentrations of the four essential amino acids measured, but less than the *Artemia*. Therefore, there were biochemical differences between the *Artemia* diet and the natural CFR diets as well as in the intestinal contents of trout fed the *Artemia* and trout fed the CFR diets. Thus, the diets are different and trout digest the diets differently.

Compared to the natural diets, Cu and Pb in the *Artemia* diet are more closely associated with the aqueous fraction. The distribution of Cu in the intestine differs with the type of food trout are fed, even if the quantity of Cu in the diets are similar (i.e., *Artemia* versus TB). Thus, while it is appropriate to compare results among fish fed the natural diets, it is



inappropriate to compare results of fish fed *Artemia* with the results of fish fed natural diets.

### Conclusions

- **Naturally contaminated diets differ from laboratory diets.** Digestion of diets fed to trout during the Woodward et al. (1994, 1995) studies differs from the digestion of diets fed to trout during the Mount et al. (1994) studies. Although Mount et al. (1994) attempted to duplicate the metal concentrations present in the Woodward et al. (1994, 1995) studies, the authors did not duplicate the natural circumstances whereby metals are metabolically incorporated into the invertebrates in the CFR. The incorporation of metals into invertebrates in the CFR occurred over time and was the result of exposures via water, diet, and sediments.
- **Partitioning of Cu and Pb differ between natural and laboratory diets.** We have documented that Cu and Pb will be distributed differently in the intestinal contents of fish fed natural diets and fish fed a diet of *Artemia* after the *Artemia* were exposed for 24-hr in the laboratory; and these differences may influence the toxicity of those diets.
- **Amino acid concentrations are different.** The amino acid concentrations in natural diets are similar to each other but are less than the concentrations in *Artemia*
- **Comparing results from studies with different diet types is improper.** It is improper to compare the results of studies performed by Woodward et al. (1994, 1995) and Mount et al. (1994). These studies used diets that are not digested in the same manner and therefore; the results are not comparable.





Figure 1. The separation of the various components of the intestinal contents collected from cutthroat trout that were fed diets of invertebrates from the CFR or diets of *Artemia* hatched in water with metals. Atomic absorption spectroscopy (AAS) was used to analyze metals in the components.

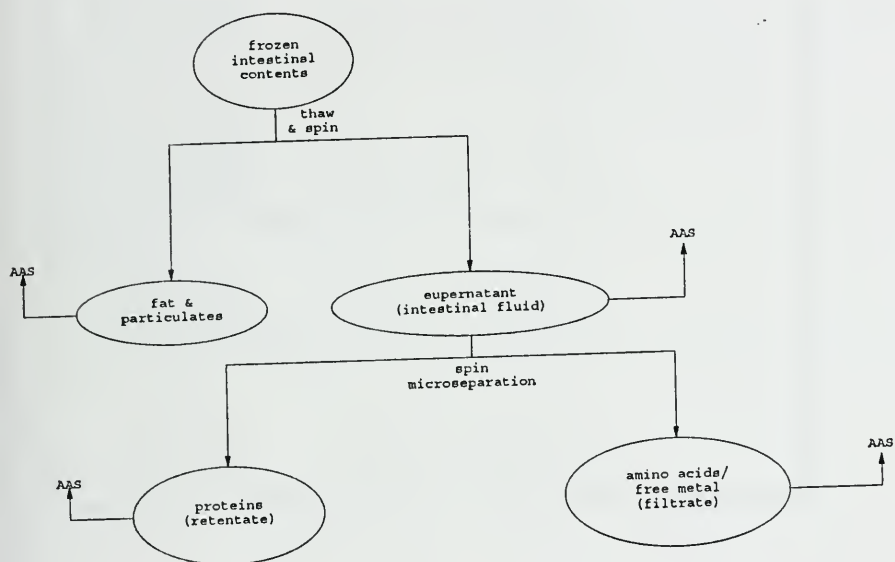




Table 1. Concentrations of amino acids measured in natural diets collected from the CFR and in *Artemia*. N = 5 unless indicated otherwise, and the SEM is in parentheses.

Amino acid Diet	Amino acid concentration ( $\mu\text{Mol/g}$ )	Percent of <i>Artemia</i> - metal
Valine		
Turah Bridge	0.133 (0.007)	60
Gold Creek	0.130 (0.031)	58
Warm Springs	0.113 (0.016)	51
<i>Artemia</i> - metal	0.223 (0.048)	100
Phenylalanine		
Turah Bridge	0.027 (0.003)	43
Gold Creek	0.014 N=1	22
Warm Springs	0.032 N=2	51
<i>Artemia</i> - metal	0.063 (0.009)	100
Leucine		
Turah Bridge	0.096 (0.004)	73
Gold Creek	0.097 (0.017)	74
Warm Springs	0.096 (0.012)	73
<i>Artemia</i> - metal	0.132 (0.030)	100
Isoleucine		
Turah Bridge	0.08 (0.007)	65
Gold Creek	0.076 (0.014)	62
Warm Springs	0.076 (0.012)	62
<i>Artemia</i> - metal	0.123 (0.033)	100



Table 2. Mean concentrations of copper (Cu) in diets ( $\mu\text{g/g}$ , dry wt) fed to cutthroat trout for 3 consecutive days, and, following the digestion of those diets, the percent of total Cu in the contents of the anterior intestine distributed between the intestinal fluids and the particulates present in the intestinal contents. N = 5 for all means, and the SEM is in parentheses. Means with different letter designations within a column are significantly different at  $p \leq 0.05$ .

Site	Diet ( $\mu\text{g/g}$ dry wt.)	Percent of total Cu in contents of anterior intestine	
		Intestinal fluid	Particulates
Turah Bridge	106 (3.9)	54 (2.7) <sup>ab</sup>	46 (2.7)
Gold Creek	188 (1.2)	44 (3.6) <sup>a</sup>	56 (3.6)
Warm Springs	181 (3.7)	47 (4.8) <sup>a</sup>	53 (4.8)
Artemia - Metals	105 (17.4)	67 (5.9) <sup>b</sup>	33 (5.9)



Table 3. Percent distribution of copper (Cu) between the amino acid/free ion fractions, and the protein fractions of intestinal fluids collected from cutthroat trout that had been fed invertebrate diets from the CFR or *Artemia*. N = 5 for all means and the SEM is in parentheses. Means with different letter designations within a column are significantly different at  $p \leq 0.05$ .

Site	Percent of total Cu in the intestinal fluid	
	Amino acid/ free metal ions	Protein
Turah Bridge	69 <sup>a</sup> (1.2)	33 (1.4)
Gold Creek	72 <sup>ab</sup> (2.1)	31 (1.5)
Warm Springs	72 <sup>ab</sup> (1.9)	26 (1.0)
Artemia - Metals	79 <sup>b</sup> (1.6)	22 (2.8)





Table 4. Mean concentrations of lead (Pb) in diets ( $\mu\text{g/g}$ , dry wt) fed to cutthroat trout for 3 consecutive days, and following the digestion of those diets, the percent of total Pb in the contents of the anterior intestine distributed between the intestinal fluids and the particulates present in the intestinal contents. N = 5 for all means, and the SEM is in parentheses. Means with different letter designations within a column are significantly different at  $p \leq 0.05$ .

Site	Diet ( $\mu\text{g/g}$ dry wt.)	Percent of total Pb in contents of anterior intestine	
		Intestinal fluid	Particulates
Turah Bridge	8 (0.4)	17 (2.0) <sup>a</sup>	83 (2.0)
Gold Creek	18 (0.2)	12 (1.1) <sup>a</sup>	88 (1.1)
Warm Springs	17 (1.9)	10 (2.3) <sup>a</sup>	90 (2.3)
Artemia - Metals	23 (3.0)	29 (4.0) <sup>b</sup>	71 (4.0)



Table 5. Mean concentrations of arsenic (As) in diets ( $\mu\text{g/g}$ , dry wt) fed to cutthroat trout for 3 consecutive days, and following the digestion of those diets, the percent of total As in the contents of the anterior intestine distributed between the intestinal fluids and the particulates present in the intestinal contents. N = 5 for all means, and the SEM is in parentheses. Means with different letter designations within a column are significantly different at  $p \leq 0.05$ .

Site	Diet ( $\mu\text{g/g}$ dry wt.)	Percent of total As in contents of anterior intestine	
		Intestinal fluid	Particulates
Turah Bridge	5 (0.3)	50 (4.1) <sup>a</sup>	50 (4.1)
Gold Creek	21 (1.4)	41 (2.9) <sup>a</sup>	59 (2.9)
Warm Springs	19 (1.4)	43 (11.7) <sup>a</sup>	57 (11.7)
Artemia - Metals	40 (4.9)	56 (7.9) <sup>a</sup>	44 (7.9)



Table 6. Mean concentrations of cadmium (Cd) in diets ( $\mu\text{g/g}$ , dry wt) fed to cutthroat trout for 3 consecutive days, and following the digestion of those diets, the percent of total Cd in the contents of the anterior intestine distributed between the intestinal fluids and the particulates present in the intestinal contents. N = 5 for all means, and the SEM is in parentheses. Means with different letter designations within a column are significantly different at  $p \leq 0.05$ .

Site	Diet ( $\mu\text{g/g}$ dry wt.)	Percent of total Cd in contents of anterior intestine	
		Intestinal fluid	Particulates
Turah Bridge	2 (0.1)	48 (4.3) <sup>a</sup>	52 (4.3)
Gold Creek	2 (0.1)	38 (5.0) <sup>a</sup>	62 (5.0)
Warm Springs	2 (0.2)	44 (6.1) <sup>a</sup>	56 (6.1)
Artemia - Metals	10 (1.1)	59 (5.6) <sup>a</sup>	41 (5.6)



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